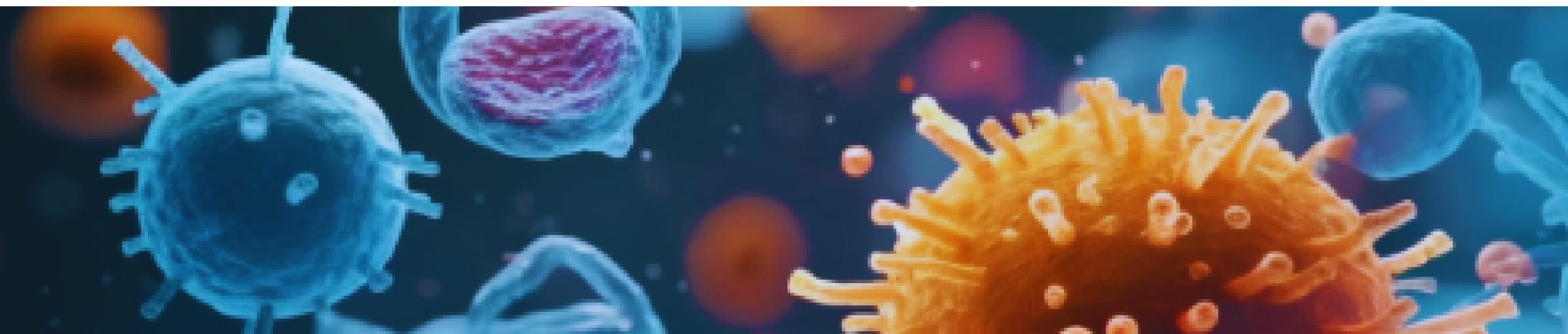


PreMiEr Research Portfolio

Creating Biologically Healthy Indoor Environments

2025



EXECUTIVE SUMMARY



PreMiEr Engineering Research Center advances precision microbiome engineering for healthier built environments—integrating biology, materials, sensing, and modeling to **reduce infection risks and promote beneficial microbial ecosystems** across healthcare and residential settings.

Monitoring & Risk analysis for Healthy Buildings

Advanced sensing and computational frameworks are transforming microbiome data into **actionable strategies for building health management**.

Lott and Brown team's Healthy Building Dashboard integrates quantitative microbial risk assessment with real-time monitoring, while *Turpin et al.* develop bioaerosol-based fungal control systems, coupling microbial inoculants with predictive ventilation modeling to guide safe, responsive air management strategies. *Noble, Brown, Callahan, and Chen* are advancing next-gen microbial surveillance, including: establishing pathogen and AMR profiles across global hospital testbeds; improving sequencing and contamination control pipelines; standardizing microbiome measurements; and deploying autonomous robotic platforms for real-time fungal source detection.

Materials Innovation for Resilient MoBEs

Materials research is redefining **how engineered surfaces and systems interact with microbial communities** to advance innovative approaches for infection prevention and the development of healthy MoBEs.

Wiesner's team develops hydrogel-based delivery systems to enable sustained release of beneficial microbes within plumbing infrastructure; *Gunsch and Anderson's group* demonstrate foam disinfectants delay pathogen colonization in hospital sinks; *Thomas and Graves' group* reveal nanoparticle geometries that prevent bacterial resistance, informing evolution-resistant antimicrobial coatings; and *Jones et al.*, reengineer plumbing designs through flow modeling and biofilm assays to limit biofilm formation.

Engineering Microbial Life for Safer Spaces

PreMiEr pioneers synthetic biology and ecological design to **precisely control and safely deploy engineered microbiomes**.

Newman's team develops modular bacteriophages to eliminate antimicrobial-resistant pathogens in plumbing systems; *Rawls' group* identifies genetic drivers of microbial persistence on built surfaces. *Delgado Vela et al.*, engineers hybrid phage-mycovirus systems targeting both bacterial and fungal pathogens, and *Crook's lab* advances H₂S-responsive genetic kill switches for biocontainment. At the community level, *Venturelli and You* group use AI-guided design to optimize microbial consortia that suppress pathogens in hospital sinks, and *Gunsch et al.* integrate antipathobiotic microbes to create resilient probiotic ecosystems that sustain healthy plumbing microbiomes.

All the engineering projects are integrated with Societal and Ethical Implication (SEI) core group, where *Kuzma and colleagues* lead societal and ethical integration, ensuring responsible innovation and public trust in microbiome engineering.

Together, these projects define an integrated framework for precision microbiome engineering—linking molecular design, microbial community dynamics, material innovations, and data-driven monitoring into a unified approach for healthier, more resilient built environments. This convergent portfolio is advancing sustainable, evidence-based strategies to **reduce healthcare-associated infections, mitigate antimicrobial resistance, and enable next-generation healthy building design that promotes beneficial microbiome functions**.

— Dr. Claudia Gunsch
Director, PreMiEr | Duke University

WHO WE ARE

PreMiEr is a National Science Foundation (NSF)-funded Engineering Research Center (ERC) headquartered at Duke University in partnership with four other universities in North Carolina.



NORTH CAROLINA
AGRICULTURAL AND TECHNICAL
STATE UNIVERSITY



THE UNIVERSITY
of NORTH CAROLINA
at CHAPEL HILL

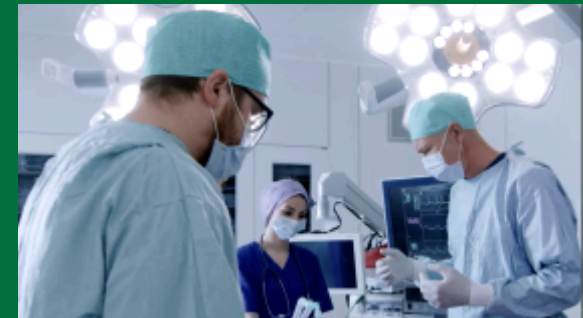
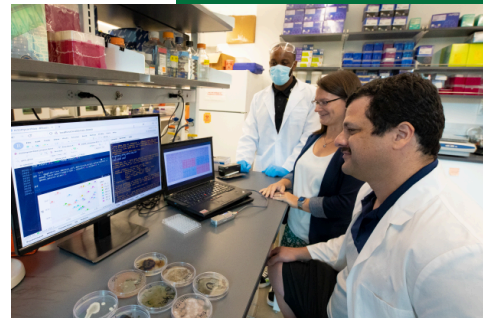


OUR VISION

To enable the bioinformed design and operation of smart, healthy built environments through microbiome engineering

OUR MISSION

To develop innovative approaches to engineer-built environment microbiomes, promoting beneficial microbes while preventing the spread of pathogens

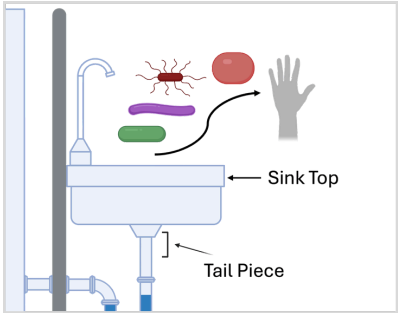


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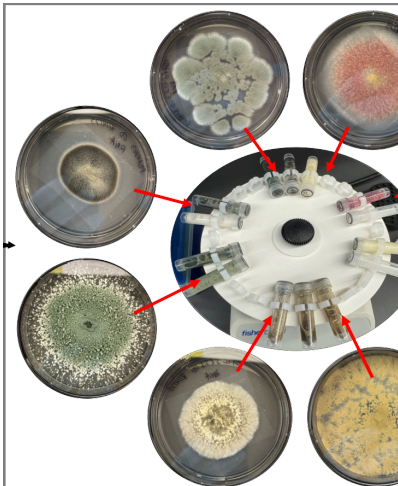
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Monitoring and Risk Modeling for Healthy Buildings

Microbial Risk Assessment to Inform Engineering Solutions in the Built Environment

Lead Faculty: Megan Lott, Anthony Fodor, Joe Brown | **Scholar:** Lindsay Saber

Problem Statement

Microbes in the built environment can both harm and benefit human health. Healthcare facilities, in particular, face challenges in managing microbial risk where infection control must balance pathogen suppression and promoting healthy microbial exposures. Despite progress in microbial monitoring, there is a lack of practical tools to integrate microbiome data into actionable health risk assessments for built environments

Project Objectives

To develop and validate **quantitative microbial risk assessment (QMRA) models** that:

- Predict and quantify health risks and benefits associated with microbial exposures in the built environment.
- Provide actionable risk thresholds and simulation tools to guide microbiome engineering interventions in healthcare settings



Research Approach

Model Development:

Construct QMRA models using metagenomic data from PreMiEr healthcare test beds.

Integration:

Combine microbial and chemical risk frameworks using common health metrics (e.g., DALYs).

Positive Dose-Response Modeling:

Establish data-driven models that capture potential health benefits of microbial exposure.

Tool Deployment:

Build a web-based risk assessment interface leveraging prior tools (e.g., MicroHibro) for interactive simulation and decision support.

Validation:

Simulate outcomes of engineering interventions such as Far UV-C disinfection and building probiotics at test sites, including Bolivian hospitals and the PreMiEr Home @ Duke

Project Goals

- Quantitative metagenomic dataset from hospital environments
- Web-based QMRA tool prototype-Dashboard
- Systematic review of microbial risk in healthcare
- Comparative analysis of chemical vs microbial risks
- Dose-response models for beneficial microbial effects

Impact

This project will establish the first integrated **risk-benefit assessment framework** for microbes in the built environment. The results will directly inform hospital design, facility hygiene strategies, and regulation of new microbiome-based interventions.

Long-term, the work will transform how microbial exposures are governed—moving from pathogen avoidance to **microbiome-informed health optimization**

Spatiotemporal Modeling and Bioaerosol-Based Control of Fungal Pathogens in Built Environments

Lead Faculty: Barb Turpin, Glenn Morrison

Scholar/Research Staff: Clara Eichler, Denene Blackwood, Sherlynette Perez Castro, Emily Nortmann, Gavin Duffy, Yanbaihui Liu

Problem Statement

Up to **20% of U.S. homes** experience mold-related issues, with fungal growth linked to **asthma and respiratory illnesses**. Traditional remediation methods focus on removal and cleaning but fail to sustain or reestablish a healthy indoor microbial ecosystem after remediation. There is a need for **sustainable, biologically driven interventions** that control fungal pathogens while preserving a beneficial indoor microbiome

Project Objective

Develop and validate an integrated microbial engineering system that uses bioaerosol delivery of beneficial inoculants and real-time sensing to monitor, suppress, and prevent fungal pathogen growth in residential and healthcare environments

Research Approach

Design and Test Biotic Inoculants:

Develop microbial cocktails that outcompete or deactivate target fungi.

Optimize Aerosolization & Dispersion:

Engineer delivery and spatial distribution through home ventilation systems using the PreMiEr Home@Duke and UNC Environmental Chambers

Implement Feedback Control:

Utilize real-time microbial volatile organic compound (MVOC) sensors and predictive spatiotemporal models for adaptive interventions.

Validation:

Conduct laboratory and real-world trials to assess efficacy and sustainability

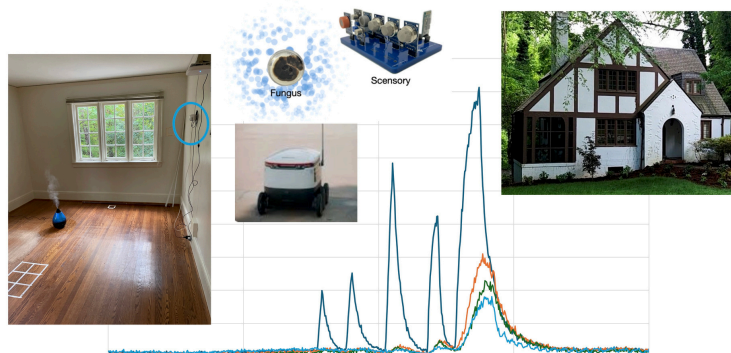
Project Goals

- Develop **optimized bioaerosol delivery** method validated by predictive models and measurements
- Further the development of a **real-time robotic MVOC sensing** system for detecting fungal markers.
- Demonstrate integration of the system to monitor and maintain a healthy microbiome with targeted delivery of inoculant when needed.

Impact

This work advances **precision indoor microbiome engineering**, offering the potential to:

- **Reduce disease transmission** through proactive fungal control.
- Enhance **microbial resilience** in homes and healthcare settings.
- Support **low-risk, scalable commercial solutions** for improving indoor air and surface quality.



Autonomous Robotics for Real-Time Fungal Source Detection in Built Environments

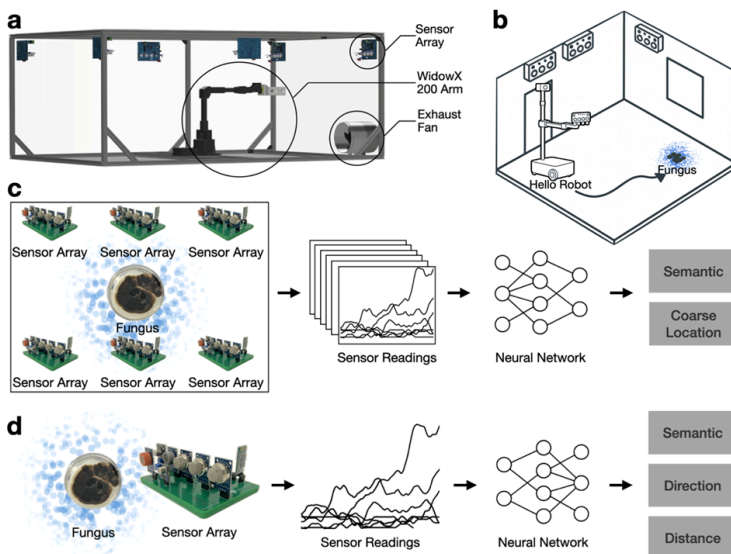
Lead Faculty: Boyuan Chen, Claudia Gunsch | **Scholar/Research Staff:** Yanbaihui Liu, Erica Babusci

Problem Statement

Fungal contamination in built environments poses **serious risks to human health, building integrity, and operational efficiency**. Current detection methods are manual, **time-consuming, error-prone, and reactive**, leading to costly remediation and health exposure. The industry needs **scalable, automated, real-time monitoring and localization solutions** to mitigate microbial contamination risks.

Project Objectives

Develop an **autonomous robotic system** that integrates **volatile organic compound (VOC) sensing, AI-driven microbial classification**, and robot navigation to detect, localize, and **monitor fungal contamination sources in real-time** across homes, hospitals, and industrial facilities.



Research Approach

VOC Sensing Array:

Designed multi-sensor and single-sensor VOC arrays for fungal species classification, spatial localization, and distance estimation.

AI & Machine Learning Models:

Developed neural networks for species recognition, contamination direction prediction, and distance regression.

Robotics Integration:

Implemented an autonomous navigation workflow combining SLAM (Simultaneous Localization and Mapping), VOC-driven sensing, and adaptive trajectory planning.

Automated Data Processing:

Developed pipelines for robust signal filtering, baseline correction, and real-time analysis.

Validation:

Conducted controlled laboratory testing and initial real-world deployment on a mobile robotic testbed.

Key Accomplishments

- Built 6 **VOC sensor arrays** capable of detecting fungal signatures with high accuracy.
- Developed **automated robotic navigation** to autonomously approach and localize contamination sources.
- Achieved real-time classification of **multiple fungal species** using AI models.
- Demonstrated **end-to-end VOC-guided navigation** on a mobile robot for source localization.

Impact

Health & Safety:

Enabling real-time monitoring of harmful microbiomes in homes and hospitals, reducing risks to human health.

Scalability:

Transitioning from manual to automated, AI-driven microbiome sampling allows large-scale adoption in diverse environments.

Industry Relevance:

Provides actionable data for healthcare, residential building management, and environmental safety industries.

Invention/Publication

Liu et al., Automated Real-Time Fungal Identification and Spatial Mapping, 2025
<https://doi.org/10.48550/arXiv.2509.19318>

Patent: Duke OTC DU8859PROV

Measurement of Residential Built Environments (BE)

Lead Faculty: Rachel Noble, Liesl Jeffers-Francis, Megan Lott, Lawrence David

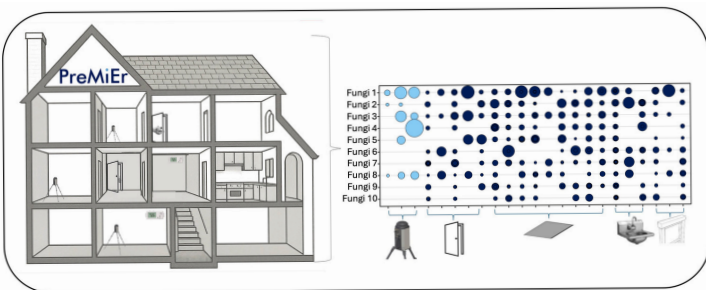
Scholar/Research Staff: Chris Cummings, Kristen Landreville, Mengyi Dong, Sherlynette Pérez Castro, Ugonna Morikwe

Problem Statement

Standardized, scalable approaches are not currently available to reproducibly quantify and assess changes in microbial targets, microbiomes, and environmental in residential and industrial BE. This **lack of standardization** hinders the ability establish thresholds or decision-based regulations to mitigate exposure-related risks, develop actionable data-driven solutions, and inform healthy and sustainable BE practices, materials, and functionalities.

Project Objectives

To advance microbial and environmental assessment methods and establish standardized, scalable approaches for characterizing the residential microbial target and microbiome, with the aim of improving indoor environmental quality, reducing harmful exposures, and protecting human health.



Schematic of the PreMiEr residential testbed showing air and surface sampling across rooms. Example fungal community profile (right) illustrates relative abundances of ten taxa across household surfaces.

Research Approach

Sampling Methods:

Real-world active and passive collection of air, surfaces (floors/walls), and premise plumbing samples using vetted, standardized, industry streamlined approaches.

Microbial Analysis:

Integration of culture-based and DNA-based assays; high-throughput sequencing including hybrid short- and long-read platforms.

Environmental Integration:

Air quality measurements and community household surveys to understand public perception and barriers to healthier environments.

Industry Engagement:

Collaboration with commercial partners (e.g., Neogen, Novonesis) to evaluate and advance products for residential and clinical applications, establishing new market opportunities.

Publications

- **Dong M. et al.** – Pipeline-specific detection of indoor mycobiomes using short- and long-read ITS sequencing approaches. (In preparation)
- **Pérez Castro S. et al.** – Bacterial diversity and occurrence of opportunistic taxa across residential kitchen surfaces. (In preparation)

Key Accomplishments

- Developed **commercially applicable tools** to measure and monitor residential BE.
- Successfully integrated **household engagement** and microbial assessments to align community perspectives with healthier practices.
- **Optimized DNA-based detection assays** (ddPCR) for major pathogens and fungi (e.g., *Aspergillus*, *Penicillium*, *E. coli*, *Klebsiella*, *Candida*).
- Advanced **microbial community assessments for both bacteria and fungal mold across households** to understand connectivity and scaling up.
- Created pathways for **new products and services** in the residential BE.

Impact

Industry:

Opens new commercial opportunities in residential health monitoring and building product development.

Healthcare & Public Health:

Improves diagnostic potential for indoor exposures influencing chronic disease, allergies, and respiratory conditions.

Community & Policy Makers:

Provides data-driven insights that inform building codes, ventilation standards, and consumer awareness efforts.

Microbiome Characterization at the Hospital Maternal Infantil–Miraflores (La Paz, Bolivia)

Lead Faculty: Joe Brown, Megan Lott | **Scholar/Research Staff:** Lindsay Saber, Ivory Blakely, Caroline Butler

Problem Statement

Healthcare-associated infections (HAIs) and antimicrobial resistance (AMR) represent a critical threat to patient safety and hospital operations worldwide. These risks are especially severe in facilities within low- and middle-income countries, where pathogen burden, differences in antimicrobial stewardship, and environmental health services present additional challenges. A baseline understanding of the built environment microbiome in hospital settings is crucial for developing strategies to mitigate infection risks.

Project Objectives

- Map the built hospital microbiome in La Paz, Bolivia.
- Identify prevalence and resistance of ESKAPEE pathogens.
- Generate applied data to guide novel hospital disinfection solutions, including Far-UV-C light.

Research Approach

Samples:
>200 from air, water, P-traps, sink and room surfaces, high-/low-touch hospital areas.

Techniques:
Microbial culture, qPCR/dPCR for pathogen identification and quantification, Illumina 16S sequencing and shotgun metagenomics for microbial community characterization, and whole-genome sequencing for resistance profiling.

Innovation:
Overcame disinfectant-related microbial growth inhibition by neutralizing buffers.

Approach

Key Accomplishments

- Characterized >159 isolates for pathogen load and antimicrobial resistance.
- Characterized >200 samples for microbial community on surfaces and in air
- Established a reproducible microbiome survey method applicable to hospitals in diverse environments.
- Created the first microbial risk profile for a Bolivian healthcare setting.

Impact

- Data-driven infection control:**
Enables hospitals to implement precision cleaning and monitoring protocols.
- Innovation enabler:**
Provides foundation for Far-UV-C disinfection trials and other **antimicrobial surface/air solutions**.
- Scalable application:**
Methods transferable to hospitals globally, particularly in high-burden, resource-constrained contexts.
- Regulatory alignment:**
Supports evidence needs for product validation and compliance in infection-prevention markets.

Advancing Sequencing Methods for Microbial Characterization of the Healthcare Built Environment

Lead Faculty: Benjamin Callahan, Josh Granek, Cynthia Gibas, Jessica Schlueter
Scholar/Research Staff: Qing Dai, Jorden Rabasco, Gerard Nasser, Rachel Rittmeyer

Problem Statement

Healthcare facilities harbor diverse microbial communities. To safely engineer and manage these environments, it is critical to **accurately identify and characterize microbes**—especially pathogens and antibiotic resistance carriers—using sequencing technologies. Existing methods face challenges with **low-biomass samples, long runtimes, and computational bottlenecks**.

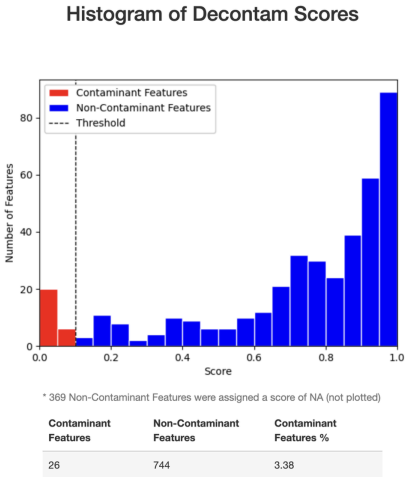
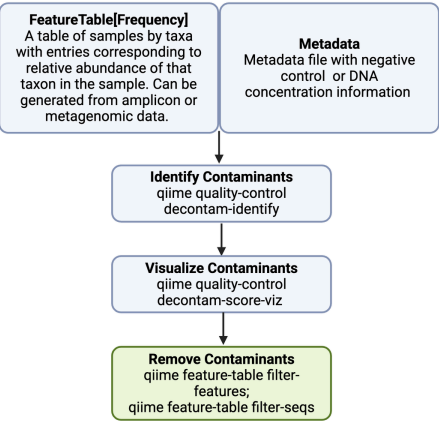
Project Objectives

Develop and validate advanced amplicon and metagenomic sequencing workflows to:

- Detect and classify microbes and pathogens
- Link antibiotic resistance genes to host bacteria
- Enhance throughput and accuracy with novel computational pipelines

Research Approach

- Generate whole genome and long-read amplicon sequences from hospital-built environments.
- Optimize sequencing & computational workflows for **low-biomass samples**.
- Improve pipelines to **remove contaminants** and **reduce runtime**.
- Validate approaches with **real hospital samples** (Duke Hospital, NC & Hospital Maternal Infantil-Miraflores, Bolivia).
- Employ **pseudo-pooling methods** to optimize sequencing accuracy and efficiency.



Key Accomplishments

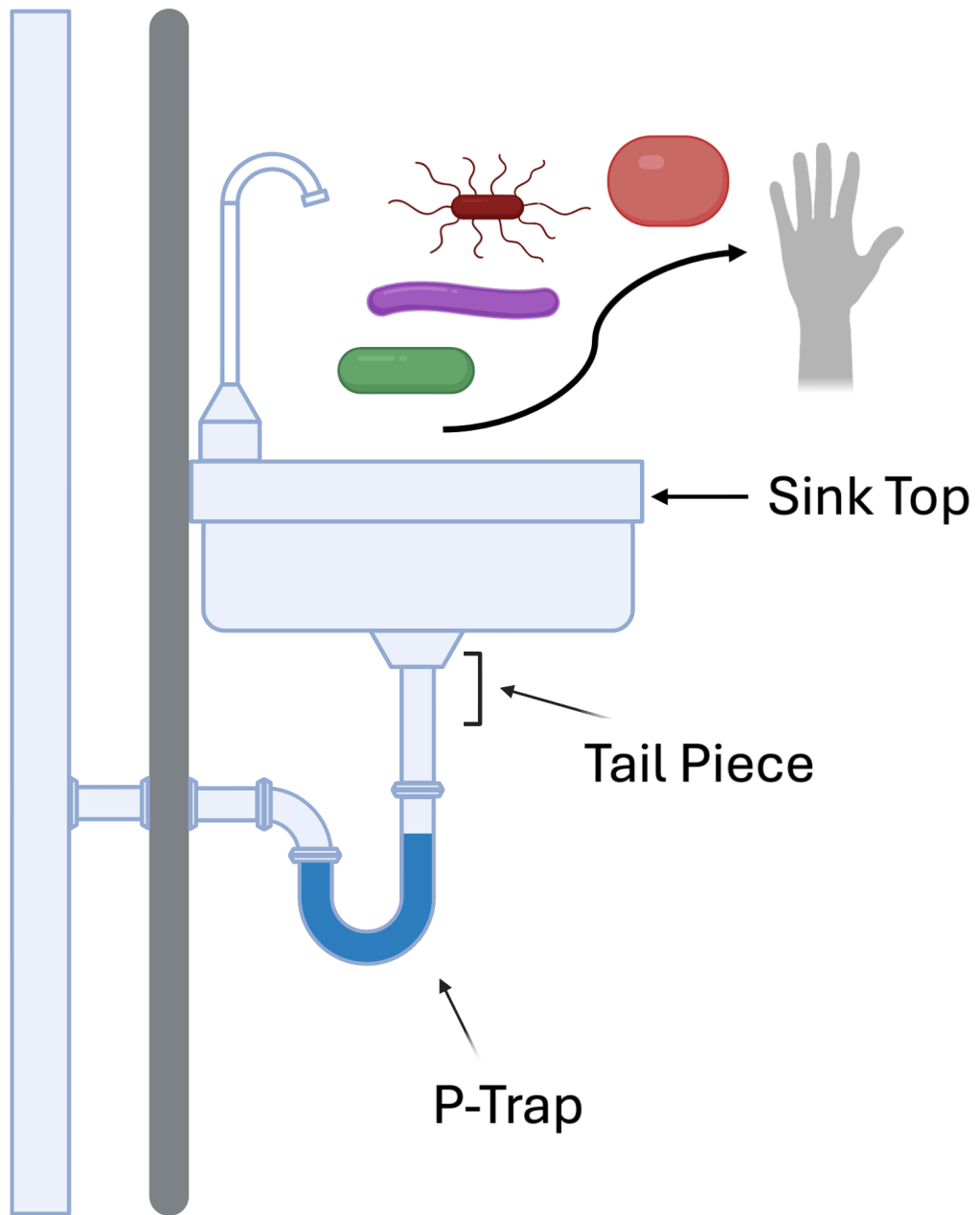
- Validated metagenome assembly workflows for Oxford Nanopore long-read shotgun sequencing.
- Integrated contaminant detection pipeline into the QIIME 2 framework.
- Developed the PseudoPooling method in DADA2 to reduce computational load.
- Benchmarked 11 commercial DNA extraction kits for assessing kitome contamination.

Impact

- This research provides **industry-ready sequencing and analysis pipelines** that:
- Increase reliability of microbiome monitoring in healthcare environments
 - Reduce costs and computational barriers to large-scale sequencing
 - Enable **early identification of pathogens and antibiotic resistance carriers**
 - Support **precision engineering of healthier, safer clinical environments**

Publication

Rabasco et al., Assessing contamination in DNA extraction kits commonly used for microbiome research
<https://www.biorxiv.org/content/10.1101/2025.09.23.677068v2>



Materials Innovation for Resilient MoBEs

Engineering Biotic Delivery to Establish Healthy and Resilient Microbiomes

Lead Faculty: Mark Wiesner | **Scholar/Research Staff:** Kobi Talma, Nathan Bossa

Problem Statement

Current techniques to disinfect plumbing systems fail to provide durable microbial control and may accelerate the development of antibiotic resistance. Biotic inoculants show promise for modulating microbial communities, but effective delivery of these inoculants is underdeveloped.

Project Objectives

- Develop a hydrogel or bio-polymer formulation that can withstand shear stresses found in hospital p-traps.
- Optimize the hydrogel or bio-polymer formulation to support the growth and establishment of biotic inoculant communities.

Research Approach

Shear Stress Resistance:

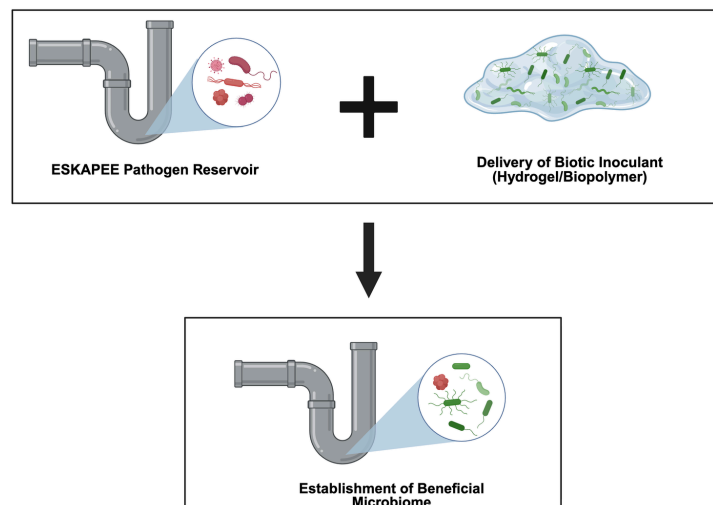
Utilize a rotating-disc rheometry approach to evaluate hydrogel/bio-polymer adhesion under fluid shear, comparing the shear stress at detachment to a threshold shear stress defined by typical values in hospital p-traps.

Biotic Inoculant Growth:

Evaluate the growth of biotic inoculants using microplate readers within hydrogel/bio-polymer and compare to growth in liquid culture.

Biotic Inoculant Establishment:

Assess the expansion of biotic inoculants from the hydrogel/bio-polymer structure into the surrounding environment using batch tests and measuring the cell concentrations in the external solution and hydrogel/bio-polymer.



Key Accomplishments

- Developed a flow-through column testing method to simulate real-world plumbing conditions.
- Demonstrated that flow conditions increase microbial attachment compared to batch tests.
- Showed that material properties alone do not strongly predict bacterial attachment, highlighting the importance of dynamic testing environments.

Impact

- Supports the development of novel delivery systems for the delivery of biotic inoculants.
- Help build a foundation for the establishment of protective microbiomes and a reduction of ESKAPEE pathogens in hospital Built Environments.

Publications

Review Paper: Material Matters:

A Framework for Integrating Surface Properties into Built Environment Microbiome Research (Submitted for publication)

Research Paper:

Evaluation of the Initial Bacterial Attachment to Built Environment Surfaces using Attachment Efficiency (α) (In preparation)

Impact of Enhanced Cleaning on Pathogens in Hospital Sinks

Lead Faculty: Claudia Gunsch, Deverick Anderson

Scholar/Research Staff: Neil Gottel, Ashlei Tomlinson, Bobby Warren

Problem Statement

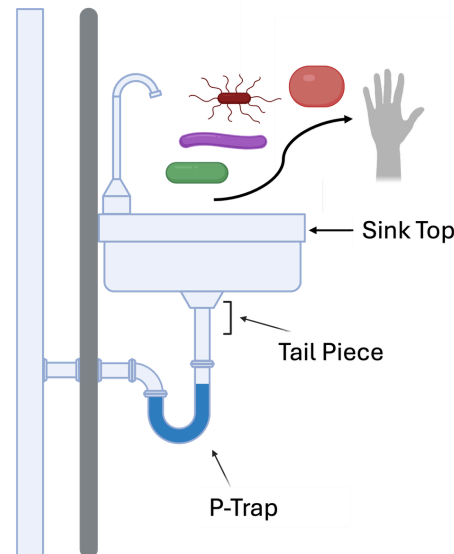
Hospital plumbing systems, particularly sinks, are frequently colonized by epidemiologically important pathogens (EIPs) such as *Acinetobacter* and *Enterobacter*. These microbes form biofilms that are difficult to remove and are linked to healthcare-associated outbreaks. Traditional cleaning remains insufficient in eliminating the risk from these EIPs, posing a risk to patients, especially if they are immunocompromised.

Project Objectives

- Assess whether the **Ecolab Virasept foaming disinfectant** improves disinfection efficacy in hospital sinks compared to traditional cleaning.
- Evaluate the impact of foam disinfectant on the **plumbing microbiome**.
- Generate **quantitative datasets** to improve risk assessment models for healthcare environments.

Research Approach

- **9-month controlled trial** in a newly built hospital unit at Duke University.
- **15 sinks treated** with foaming hydrogen peroxide disinfectant, **15 untreated controls**.
- Weekly sampling of sinks (drains, tailpiece, P-trap): 2,500 samples processed for **16S rRNA sequencing and culturing**.
- Rigorous contamination controls for sequencing reliability.



Key Accomplishments

- **Delayed colonization of EIPs by 8 weeks** with foaming disinfectant.
- Colonization after 11 weeks: **80% in control sinks vs. 20% in treated sinks**.
- **Detected 498 EIPs** in total: 433 from controls, 65 from treated sinks.
- Identified overlap between **patient bacteriological cultures** and sink isolates, confirming sinks as a reservoir
- **Only 9% of intervention sink samples were positive for EIPs**, compared to 270 (47%) in control sinks.

Impact

- Demonstrates the feasibility of **foam-based disinfectants** as superior interventions for controlling sink-borne pathogens.
- **Significantly reduces outbreak risk** from hospital plumbing reservoirs.
- Findings support **integration into infection prevention protocols**, reducing healthcare-associated infections (HAIs) and associated costs.

Investigating the Role of Nanoparticle Geometry in Preventing the Evolution of Bacterial Resistance

Lead Faculty: Misty Thomas, Rachel Bangle, and Joseph L. Graves Jr.

Scholar//Research Staff: Jacqueline Joseph-Lainez, Arrington Ashford, Leah Worrill, Muzan Ebrahim, Kennedy Whitley, and Absa Aw

Problem Statement

The escalation of multidrug-resistant microorganisms—especially ESKAPEE pathogens—poses a severe threat to global health. Their biofilm-forming capabilities enable them to resist traditional disinfection and antimicrobial treatments, leading to higher healthcare costs and infection risks. High-touch hospital surfaces such as bed rails and tray tables act as persistent microbial reservoirs. Current disinfectants are short-lived and may unintentionally accelerate resistance, creating an urgent need for next-generation antimicrobial surfaces that provide **durable, evolution-resistant, and environmentally safe protection**

Project Objectives

- Develop and validate **antimicrobial surfaces** integrating **nanoparticle** and bioinspired design strategies.
- Demonstrate **broad-spectrum antimicrobial efficacy** and **biocompatibility** in clinically relevant environments.
- Assess **long-term durability** and resistance to microbial adaptation through controlled evolutionary studies

Research Approach

Surface Development:

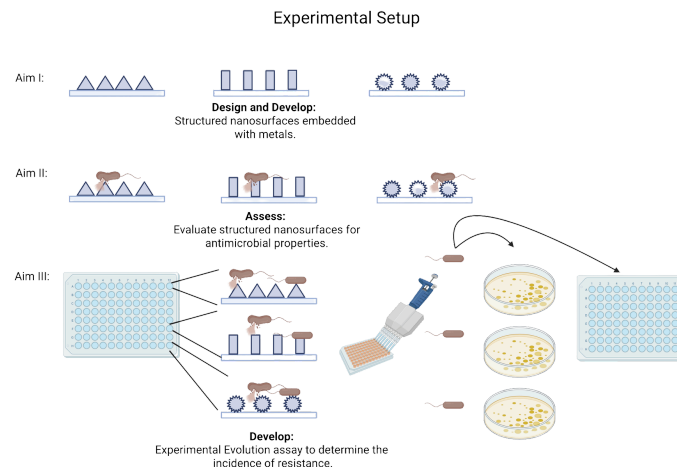
Engineer and optimize nanoparticle-based and bioinspired coatings designed for healthcare materials.

Performance Testing:

Characterize antimicrobial activity, biofilm suppression, and safety performance under laboratory conditions.

Durability Assessment:

Employ experimental evolution to evaluate microbial adaptation potential and ensure long-term stability of the antimicrobial effect



Key Accomplishments

Resistance Studies:

Identified that bacteria can evolve resistance to silver nanowires; genomic sequencing unraveled the mechanisms behind this adaptation.

Comparative Findings:

Demonstrated that silver nanoplates pose a greater challenge for microbes to develop resistance, suggesting geometric factors are key; however, commercial discontinuation limited extended testing.

Translational Insight:

Findings informed the next phase of development, emphasizing multi-edged nanoparticle geometries as a promising avenue for sustained antimicrobial function

Impact

Innovation in Antimicrobial Surfaces:

Pioneers advanced coatings offering long-lasting passive pathogen control for healthcare and high-touch environments.

Resistance Mitigation Framework:

Introduces an evolution-informed approach to material design, directly addressing antimicrobial resistance and biofilm-related failures.

Industry and Clinical Readiness:

Delivers data and prototypes that strengthen commercial partnerships with surface coating manufacturers and medical device developers, advancing deployment readiness

Interventions to limit microbial colonization in hospital plumbing systems

Lead Faculty: Andrew Jones

Scholar/Research Staff: Kayla Fericy, Andrew Wrench, Chandler Wimmer

Problem Statement

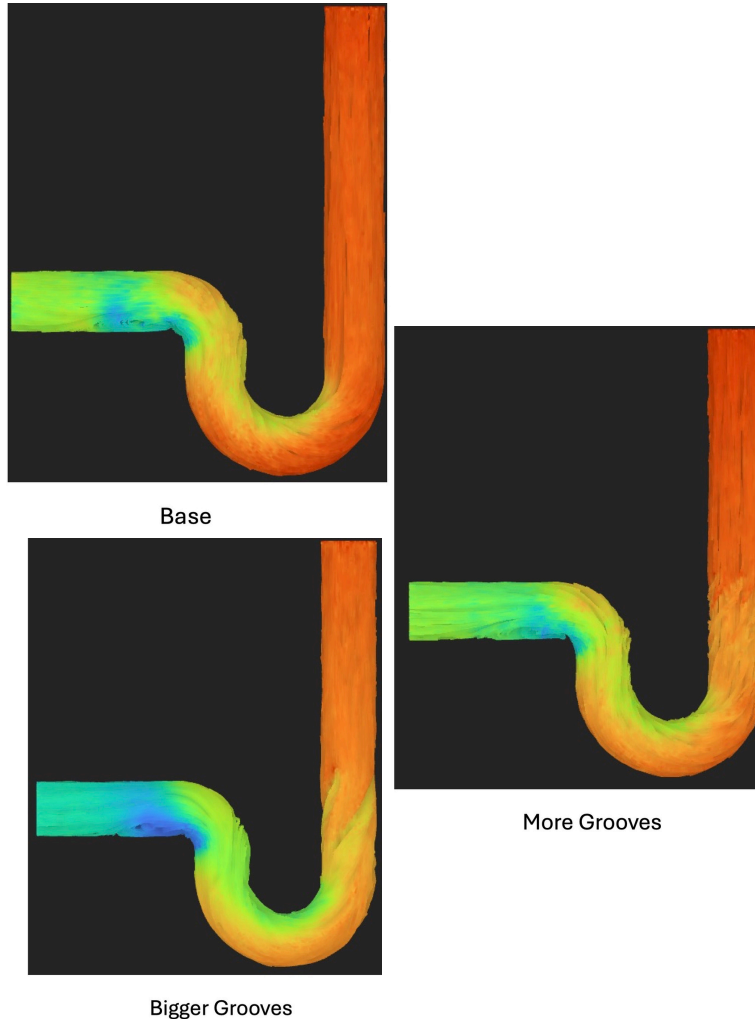
Hospital plumbing systems harbor microbial communities, potentially including ESKAPE pathogens, which contribute to hospital-acquired infections. Current plumbing designs facilitate microbial colonization and biofilm formation in components like sink p-traps, posing infection risks while maintaining critical system functionality.

Project Objectives

- Create and model flow environments inside a p-trap to study geometric impacts.
- Validate biofilm formation and growth experimentally in engineered systems.

Research Approach

- Modeling fluid flow in modified p-trap geometries to explore microbial removal.
- Fabrication and testing of modified p-trap designs to reduce pathogen persistence.



Simulation of pressure in a commercially available and bio-inspired p-trap

Key Accomplishments

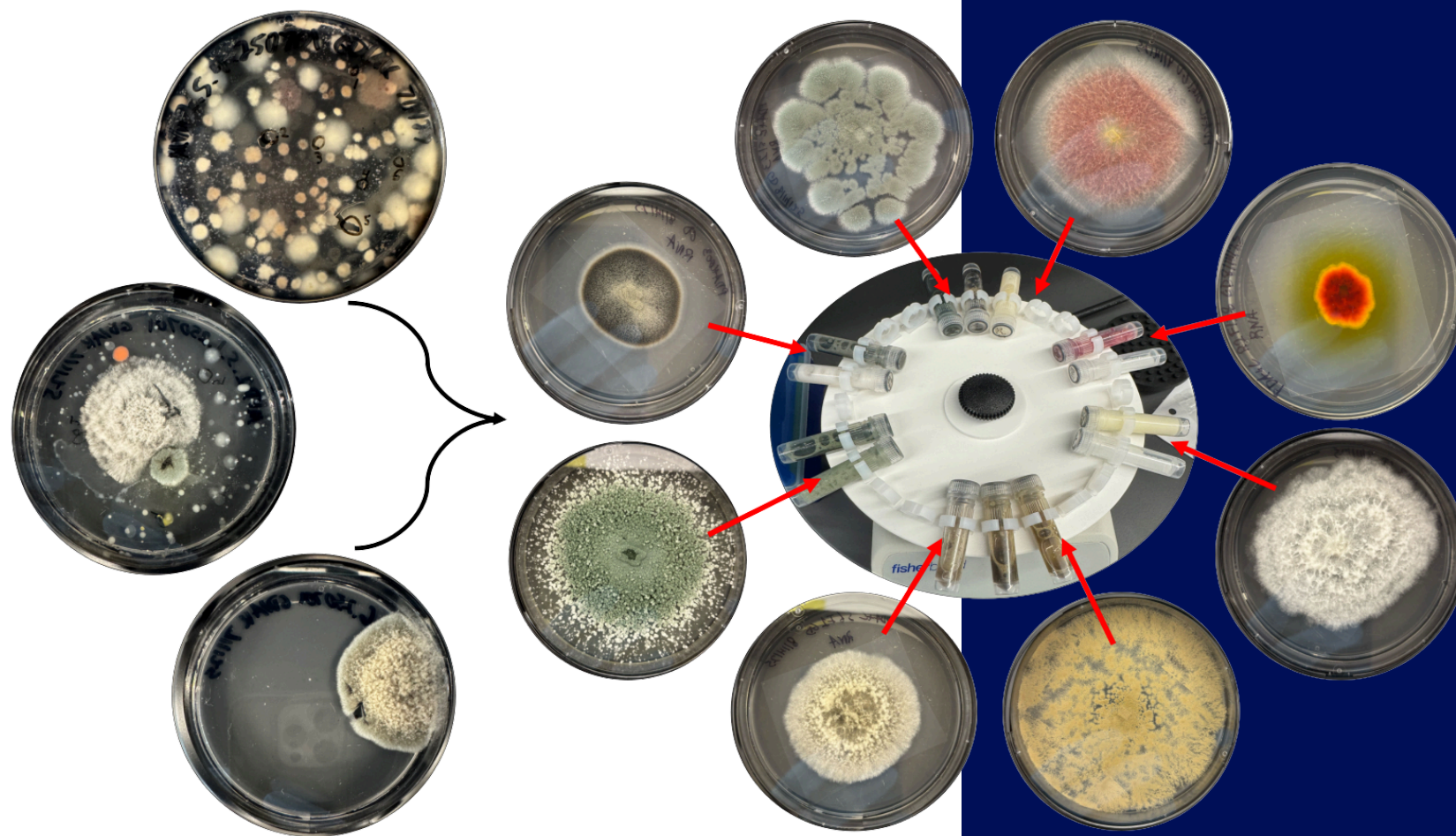
- Identified practical design intervention: p-trap covers for ADA §606.5 compliance.
- Filed patent for a standardized antibiofilm test method, already optioned for license.

Impact

- Supports redesign of plumbing infrastructure in healthcare facilities to reduce biofilm formation.
- Reduces hospital-acquired infections, leading to **lower healthcare costs and improved patient safety**.

Invention

Patent: “Plate Assay Device And Method For In Situ Imaging” – Application #18/411,564 (Filed Jan 2024, Published Jul 2024).



**Engineering Microbial
Life for Safer Spaces**

Synthetic Biology for Bacteriophage Construction

Lead Faculty: Robert H. Newman

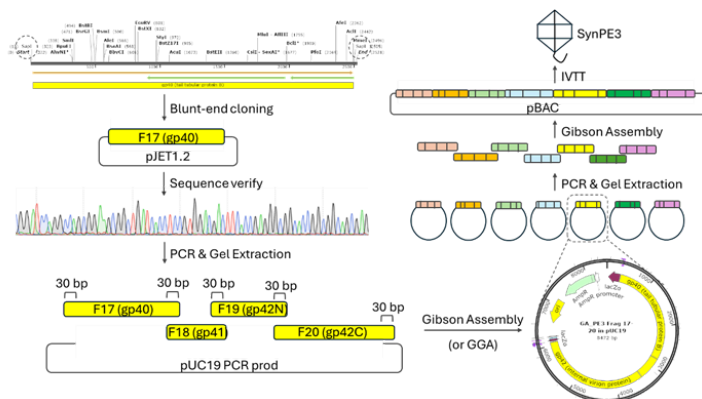
Scholar/Research Staff: Akinwunmi "Rapheal" Afuape, Madison Pinkeney, Tayler Scott, Nhoaa Powell, Adam Yang, Jaden Gilliam, Penelope Stewart

Problem Statement

ESKAPEE pathogens pose a major threat in healthcare and built environments by spreading antimicrobial resistance (AMR). They colonize plumbing systems and increase patient risk. Traditional methods lack effective long-term control. Synthetic bacteriophages have not been deployed against these pathogens in plumbing systems, leaving an urgent need for novel, targeted biocontrol solutions.

Project Objectives

- Develop a generalizable workflow for synthetic phage genome assembly and phage production.
- Engineer bacteriophages capable of targeting AMR pathogens, while incorporating genetic elements that delay, or even inhibit, pathogen resistance to phage infection.



Workflow for construction of a synthetic variant of the *P. aeruginosa* PE3 phage (SynPE3).

Research Approach

Genome engineering strategies:

Gibson Assembly, Golden Gate Assembly, bacteriophage recombineering using electroporated DNA (BRED).

De novo gene synthesis:

Building phages “from the ground up” via modular genome fragments.

Genetic optimization:

Incorporation of SapI restriction sites, reassignment of stop codons, and custom substitutions for enhanced functionality.

Application:

Designer phages for targeted elimination or modulation of plumbing system microbiomes.

Key Accomplishments

- Constructed a **modular synthetic PE3 bacteriophage genome**.
- Developed **mini-chunk assembly strategy** enabling rapid recombination of gene sets.
- Introduced non-canonical features (restriction sites, codon recoding) for enhanced modularity and gene editing.
- Successfully assembled 26 of the 28 gene fragments into their corresponding mini-chunk and constructed 5 of the 7 full-length mini-chunks needed for synthetic phage genome assembly.

Impact

Healthcare & Infrastructure Safety:

Provides a versatile and sustainable strategy to remove AMR pathogens from built environments, reducing healthcare-associated infections.

Industry Relevance:

Enables scalable synthetic biology methods for antimicrobial control in hospitals, water systems, and biotechnology facilities.

Technology Advancement:

Establishes a platform for rapid bacteriophage engineering, adaptable to multiple industrial and health applications.

Fitness Determinants and Functional Modulation in *Phocaeicola vulgatus*

Lead Faculty: John Rawls

Scholar/Research Staff: Jessica McCann, Ethan Rehder, Yelizaveta Rassadkina, Megan DiFranco, Lila Teitle

Problem Statement

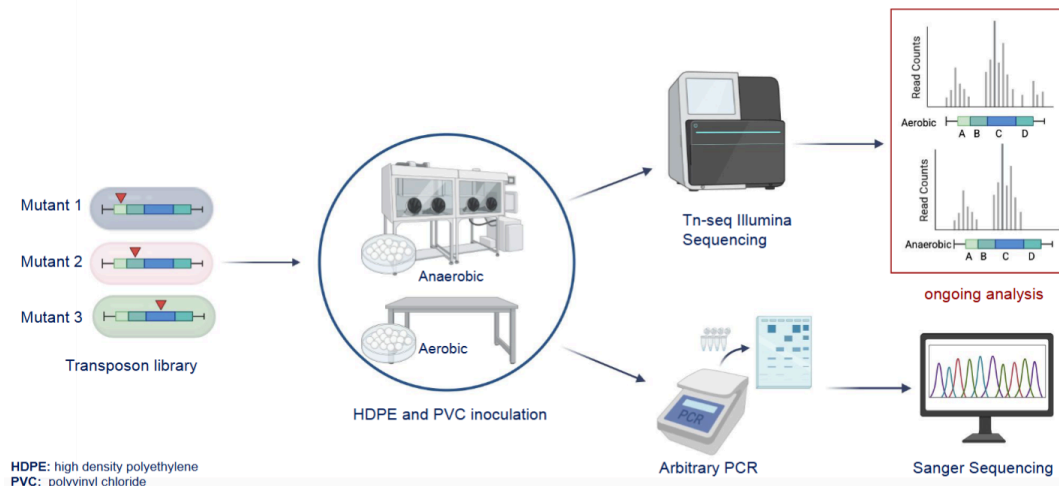
Understanding microbial community function is central to microbiome science. A major challenge lies in identifying the genetic determinants that shape microbial fitness and interactions within built environments (BEs). This project uses *Phocaeicola vulgatus* (*Pvu*), a model bacterium, to decipher how genetic variation drives survival and adaptation on BE surfaces.

Project Objectives

- Identify genes in *Pvu* required for fitness on different built environment surfaces.
- Develop technologies for functional modulation and in situ localization of *Pvu*.

Research Approach

- Application of **transposon insertion sequencing (Tn-Seq)** to map genes essential for BE surface survival.
- Development of **custom BE surface platforms conducive** to microbial colonization studies.
- Engineering tools for **functional modulation, gene/protein tagging, and localization** in *Pvu*.
- Iterative BE experiment designs with increased scale and replicates to refine datasets.



Key Accomplishments

- Finalized a robust BE surface experimental platform for Tn-Seq studies.
- Completed first full replicate of *Pvu* Tn-Seq study across **two different BE surface materials**.
- Developed **new transposon mutagenesis tools** with antibiotic resistance markers for broader applications.
- Optimized **protein tagging and isolation methods** in *Pvu*.

Impact

Scientific:

Establishes a mechanistic understanding of microbial adaptation on BE surfaces.

Technological:

Provides new tools for microbiome functional engineering.

Industry Relevance:

Lays the groundwork for applications in indoor environmental microbiome control, biofilm management, and engineered living surfaces.

Bioengineered Viral Tools for Controlling Pathogens in Residential Environments

Lead Faculty: Jeseth Delgado Vela, Nicole Rockey, Robert Newman, Kevin Garcia

Scholar/Research Staff: Gavin Duffy, Crissy Massimino, Akinwunmi (Raphael) Afuape, Brianna Jamison, Madison Pinkney, Tayler Scott

Problem Statement

The residential built environment harbors bacterial and fungal communities, including harmful ESKAPEE pathogens (*E. coli*, *Klebsiella*, *Pseudomonas*, etc.) and fungal molds. Existing antimicrobial approaches are ineffective. There is a need for novel, biologically based strategies to regulate these microbial populations.

Project Objectives

- Develop engineered bacteriophages and mycoviruses to target and control pathogenic microbes in residential environments.
- Establish viral-host systems and engineer quorum-sensing (QS) response systems in phages to disrupt biofilms and pathogen persistence.
- Develop fungal suppression mechanisms via mycoviral application.

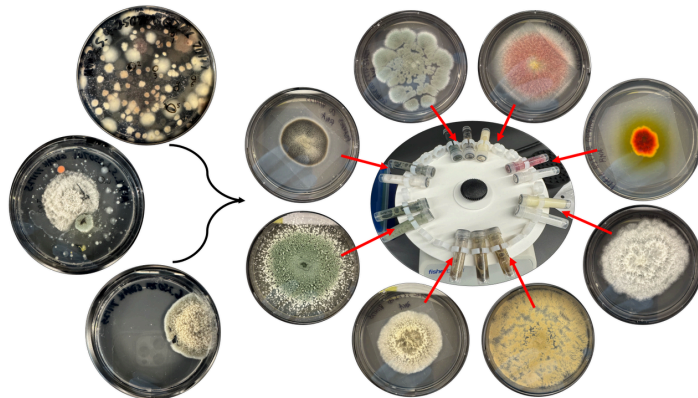
Research Approach

Mycovirus Engineering:

- Isolate fungal hosts and mycoviruses via culture-dependent/-independent methods.
- Transform mycovirus transcripts into fungal hosts and evaluate the efficacy of fungal suppression.

Phage Engineering:

- Engineer bacteriophages with QS-receptor genes to trigger pathogen lysis.
- Apply whole-genome synthetic biology approaches for phage design.
- Develop phage delivery methods for the built environment.



Key Accomplishments

- Conducted pilot sampling of 51 built-environment samples (air, surfaces, water).
- Isolated 28 fungal strains for DNA/RNA sequencing.
- Successfully isolated 4 novel *Klebsiella* phages from wastewater, 2 of which lysed all 9 tested isolates.
- Designed synthetic phage PE3 for *Pseudomonas aeruginosa* based on modular DNA fragments.

Impact

- Introduces biological alternatives to chemical disinfectants.
- Provides a scalable platform for pathogen-specific biocontrol in homes and critical built environments.
- Lays groundwork for synthetic phage therapies addressing antimicrobial resistance.

Kill Switches for Biocontainment

Lead Faculty: Nathan Crook

Scholar: Qiaochu Li

Problem Statement

The safe deployment of genetically modified microorganisms (GMOs) in built environments is hindered by the risk of unintended environmental release. Traditional containment systems lack selectivity and efficiency, limiting their real-world application.

Project Objective

To develop a **hydrogen sulfide (H₂S)-mediated** kill-switch system in *E. coli* that ensures selective, efficient, and reliable microbial cell death in wastewater environments, thereby minimizing biosafety risks while maintaining system performance.

Research Approach

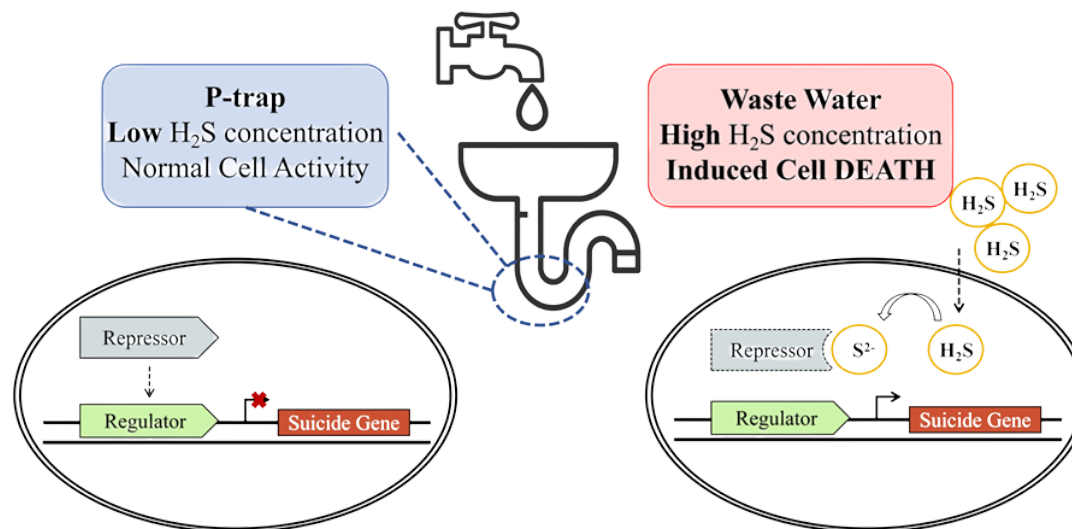
- Construct H₂S-responsive kill switch system in *E. coli*.
- Optimize switching sensitivity based on H₂S concentrations in plumbing P-traps.
- Engineer and monitor the system within defined microbiome communities.

Key Accomplishments

- Cloned and validated H₂S-responsive operator, inhibitor, and killer enzyme genes.
- Constructed inhibitor-mCherry fusion protein and verified expression.
- Designed operator sequence driving reporter expression and demonstrated inhibition in preliminary data.

Impact

- Provides a scalable, selective biocontainment solution supporting safe deployment of engineered microbes in real-world infrastructure.
- Reduces risk of GMO escape into the environment.
- Enables broader industrial adoption of microbiome engineering with enhanced biosafety compliance.



P-trap Biotic Inoculant for Inhibiting Pathogen Colonization in Plumbing Systems

Lead Faculty: Claudia Gunsch, Ophelia Venturelli, Lingchong You,

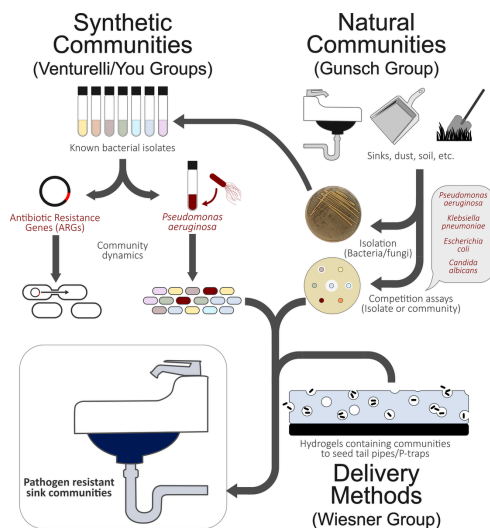
Scholar/Research Staff: Will Stiffler, Qing Dai, Neil Gottel, Aaron Yip, Conrad Moss, Zhenqqing Zhou, Kristen Lok, David Singleton

Problem Statement

Plumbing systems can harbor and disperse pathogens into buildings, posing serious health risks. Current disinfection methods are insufficient, and resistant microbial communities often persist. There is a need for innovative microbial solutions in developing conditions and inocula that result in stable, healthy microbiomes resilient to colonization by pathogens.

Project Objective

Develop, test, and optimize biotic inoculants (antipathobiotics) that exclude pathogens in plumbing systems, ensuring long-term stability and resilience through eco-friendly, deliverable, and scalable approaches.



Research Approach

Isolation & Screening:

Identify bacterial and fungal strains with antipathobiotic functions from residential and hospital P-trap samples.

Pathogen Screening:

Test against ESKAPEE pathogens, *Legionella pneumophila*, and *Candida* strains.

Functional Analysis:

Characterize mechanisms of pathogen exclusion in mixed microbial communities.

Delivery Optimization:

Introduce antipathobiotics using encapsulation, hydrogels, or other delivery/colonization methods to enhance survival and long-term efficacy.

Modeling & Testing:

Validate with laboratory-built P-trap reactors and predictive modeling.

Key Accomplishments

- Isolated a *Bacillus velezensis* strain with antipathobiotic surfactant-producing capabilities.
- Confirmed strain viability post-encapsulation.
- Constructed and validated a mock P-trap experimental reactor.
- Collected and analyzed **168 residential P-trap samples** for long-read PacBio sequencing.
- Demonstrated suppression of *E. coli* O157:H7 in several tested P-traps.

Impact

Public Health Benefit:

Reduces colonization and transmission of dangerous pathogens (*Legionella*, ESKAPEE pathogens, *Candida*).

Sustainable Solution:

Establishes eco-friendly microbial consortia with long-term stability.

Industry Application:

Provides a scalable microbial-based technology for safer building water systems.

Regulatory & Market Relevance:

Supports emerging demand for next-generation disinfection and biofilm control solutions in healthcare, residential, and commercial plumbing contexts.

Measuring and Modeling Sink Communities Using Mechanistic Modeling and Machine Learning

Lead Faculty: Lingchong You

Scholar/Research Staff: Kristen Lok, Zhengqing Zhou, Katie Duncker & Emrah Simsek

Problem Statement

Hospital sinks serve as reservoirs for antibiotic-resistant and pathogenic bacteria that can spread to patients and cause hospital-acquired infections (HAIs). HAIs are responsible for **90,000 deaths annually** and cost **\$30–45 billion per year** in the U.S. Current monitoring methods are inefficient, limiting the ability to detect, predict, and control microbial risks in hospital sink environments.

Project Objectives

- Develop engineered microbial sentinels to detect pathogen-associated chemicals in sink drains.
- Create a **synthetic sink community** to study antibiotic resistance and pathogen dynamics.
- Build an **AI-driven measurement** tool to efficiently track sink community changes from morphology.

Research Approach

- Construct **synthetic microbial consortia** to sense pharmaceuticals and antibiotics in sink P-traps.
- Use **machine learning pipelines** (variational autoencoder + MLP) to analyze fluorescence and colony morphology.
- Isolate and characterize sink bacteria by **genomic sequencing, phenotyping, and high-throughput assays**.
- Predict and model microbial growth, antibiotic resistance, and interactions under controlled conditions.

Key Accomplishments

- Developed **SynkC**, a synthetic sink community, to demonstrate keystone species driving pathogen expansion under antibiotic stress.
- Created an **ML pipeline** to measure microbial abundances in sink isolates with high accuracy ($R^2 > 0.8$ – 0.95).
- Produced synthetic microbial sensors capable of detecting chemical compounds and predicting community dynamics.

Impact

- Enables faster diagnosis of **nosocomial (hospital-acquired) co-infections**.
- Provides **low-cost, scalable tools** for monitoring and mitigating microbial risk in healthcare facilities.
- Supports **next-generation infection control strategies**—reducing healthcare costs and mortality.
- Broad applicability to **industrial, clinical, and environmental microbiome monitoring**.

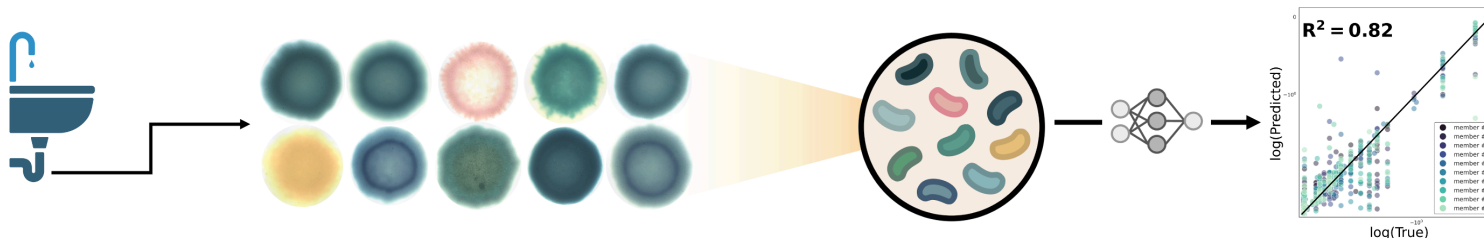
Publications

bioRxiv Preprint (2025):

“Keystone engineering enables collective range expansion in microbial communities”

bioRxiv Preprint (2025):

“Engineering microbial consortia for distributed signal processing”



Design and optimization of synthetic microbial communities to inhibit pathogens in the built environment

Lead Faculty: Ophelia Venturelli

Scholar/Research Staff: Aaron Yip, Conrad Moss

Problem Statement

Hospital sinks act as reservoirs for microbial communities that harbor pathogenic bacteria and antibiotic resistance genes. Pathogen persistence on such surfaces increases risks of infections and accelerates the spread of antibiotic resistance, posing a critical challenge for healthcare environments.

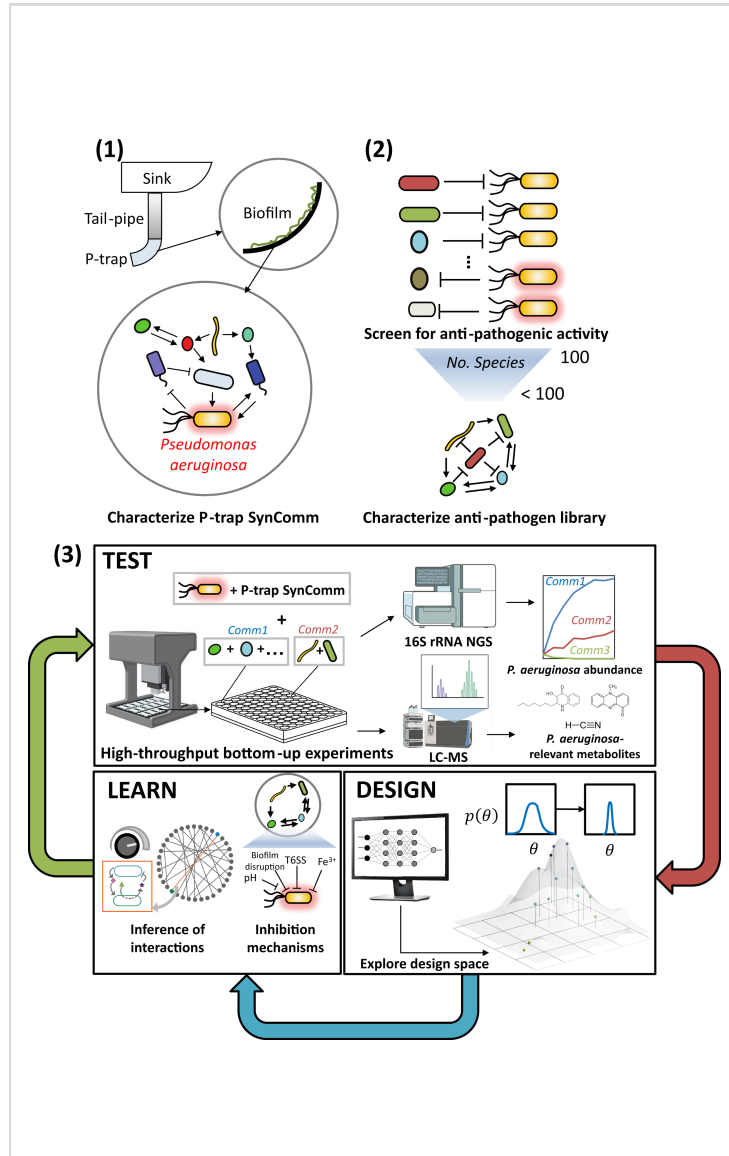
Project Objectives

Engineer stable, robust microbial communities that:

- Inhibit the growth of *Pseudomonas aeruginosa* (target pathogen).
- Suppress biofilm formation.
- Block the spread of antibiotic resistance genes.

Research Approach

- Characterize the growth and pathogen inhibition potential of microbial species from soil and the built environment under controlled conditions.
- Develop high-throughput assays to measure:
 - Biofilm production
 - Plasmid persistence
 - Pathogen abundance
 - Species abundance in microbial communities
- Integrate experimental data with ML, mechanistic computational modeling, and multi-objective optimization to predict community dynamics.
- Apply active learning to optimize microbial community performance against pathogens.



Key Accomplishments

- Screening >100 diverse bacteria for pathogen inhibition potential using a synthetic community of the hospital sink
- Developed physics-informed computational frameworks to predict community and plasmid dynamics.
- Apply active learning techniques to guide large experimental design landscapes to optimize the desired functions of microbial communities
- Created scalable experimental measurements for quantifying biofilm and plasmid persistence

Impact

Healthcare:

Reduces infection risks and antibiotic resistance spread in hospital settings.

Technology:

Provides predictive modeling frameworks combining microbiology, machine learning, and synthetic community design.

Industry Application:

Potential for deployment in hospital sinks, water systems, and infection control infrastructure

Societal and Ethical Implications of Microbiome Engineering

Lead Faculty: Jennifer Kuzma, Joe Brown, Joseph Graves Jr., Jean Cadigan

Scholar/Research Staff: Kristen Landreville, Chris Cummings, Nourou Barry, Darcy Anderson, Rich Eva

Problem Statement

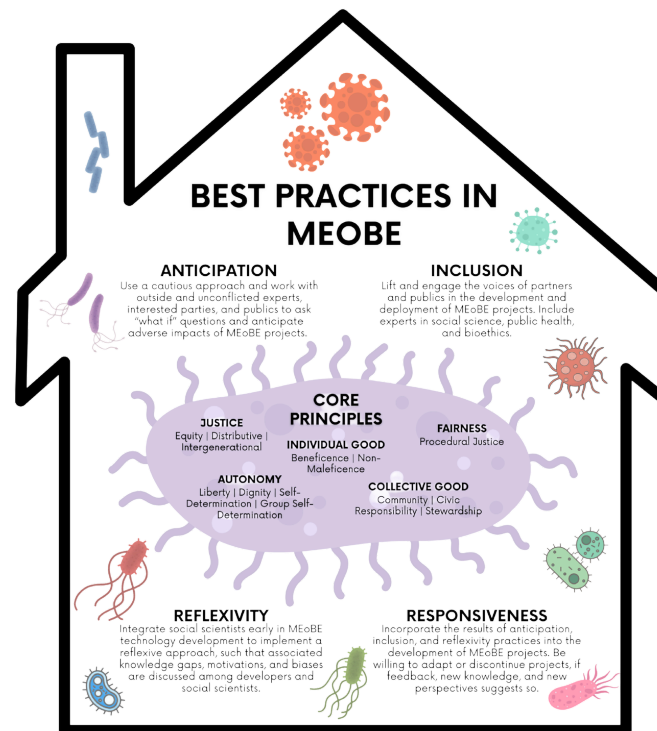
Microbiome engineering offers significant opportunities for improving BE (e.g., reducing mold, tackling antimicrobial resistance). However, public trust, societal acceptance, and ethical oversight are critical hurdles. Without clear principles for responsible development and integration of societal perspectives, public resistance and policy challenges may slow down innovation.

Project Objectives

- Establish principles & practices for responsible microbiome engineering in BE.
- Integrate societal, ethical, and policy insights within scientific and engineering projects.
- Evaluate public perceptions, hopes, and concerns regarding microbiome engineering.
- Assess public communications on microbiome studies and its influence on research and policy.

Research Approach

- **International Deliberative Workshops:** Co-developed principles for responsible innovation.
- **Stakeholder & Community Engagement:** Conducted surveys and interviews on real-world cases (e.g., household mold, hospital AMR).
- **National Quantitative Survey:** Measured U.S. public attitudes and key demographic drivers.
- **Textual Content Analysis:** Analyzed public communication of microbiome research and its impact on trust in science.



Key Accomplishments

- Developed and validated principles for the responsible microbiome engineering.
- Published two national peer-reviewed studies on U.S. public attitudes
- Identified key acceptance factors: demographics, values, and knowledge.
- Designed and implemented surveys and interviews on real-world interventions.
- Organized international workshops for scientists, industry, and policymakers.

Impact

- Establishes a framework for responsible microbiome innovation in industry.
- Offers insights into public perception to inform product and regulatory strategies.
- Fosters ethical alignment among industry, policymakers, and communities.
- Promotes trust, acceptance, and responsible scaling of BE microbiome technologies.

Publications

Cummings CL et al., (2025) Public perceptions and support for introduced microbes to combat hospital-acquired infections and antimicrobial resistance. *PLoS One* 20(10): e0332578.

Cummings, C. et al., (2025). Natural vs. Genetically Engineered Microbiomes: Public Attitudes and Pathways for Future Engagement. *Frontiers in Genetics* 16.

Other Publications in 2024

Cummings, C. *et al.*, *Frontiers in Public Health*. 12:1477377

Hardwick A et al, *Environment, Systems and Decisions* 44: 215–238.

CURRENT TEST BEDS

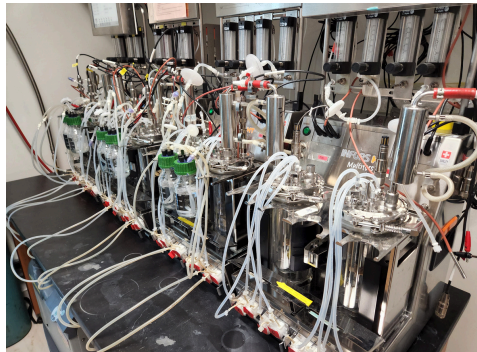
ENVIRONMENTAL CHAMBERS

Precision-controlled environmental chambers for studying aerosolized microbial spread, colonization, and interactions with building materials.



ENVIRONMENTAL BIOREACTORS

Environmental bioreactor testbeds that simulate engineered indoor built environments to design and test microbiome interventions



PREMIER HOME @ DUKE

Single family home residential testbed for conducting MoBE research and tech validation.



PARTNER HOSPITALS

Hospital test beds across North Carolina and South America for conducting MoBE research and tech validation.



GLOBAL COMMUNITY SITES

Collaborations with communities in Bolivia and both urban & rural North Carolina for field-based research.



PRINCIPAL INVESTIGATORS



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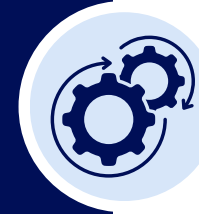
Collaboration

Joint funding opportunities for external grants



Financial

Leverage R&D funds through shared costs and significantly reduced overhead



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