BioSciences

BER Pilot Project

Advanced Biorecovery of Critical Minerals through AI/ML-Guided Design of Microbial Chassis

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U.S. DEPARTMENT OF ENERGY

BERKELEY LAB

Funding and resources

- Total: $850K for FY26 (funds received end of August), subject to renewal in fall 2026
- IUT between LBNL and UC Berkeley has been established, MTA in progress
- Molecular Foundry user proposal has been submitted to leverage advanced imaging capabilities

Team Member	Role	Division	Effort	Budget
Ning Sun	PI	BSE / ABPDU	10%	$ 264,870
Rita Kuo	Research Support		20%	
Saad Naseem	Postdoc		50%	
Norma Cecilia Martinez-Gomez	Co-PI	UC Berkeley	10%	$ 158,599
Trinity Reiner	Lab Technician		80%	
Yasuo Yoshikuni	Co-PI	JGI	5%	
Yusuke Otani	Postdoc		50%	$ 137,622
Rebecca Abergel	Co-PI	Chemical Sciences	5%	
Alexander Brown	Postdoc		50%	$ 130,254
Marcin Joachimiak	Co-PI	EGSB	10%	
Mark Miller	Software Developer		20%	$ 131,682
Romy Chakraborty	Co-PI	EESA	2%	
Mingfei Chen	Postdoc		5%	$ 26,973
TOTAL				**$ 850,000**

BER Decision Letter

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BER is most interested in supporting LBNL objectives that seek to build a high-throughput strain screening and validation pipeline for ML guided bioengineering of REE accumulation, as these aims are more directly relevant to the request in the call to advance biodesign and synthetic biology for the extraction and recovery of CMM from natural and complex environments.

Project Overview

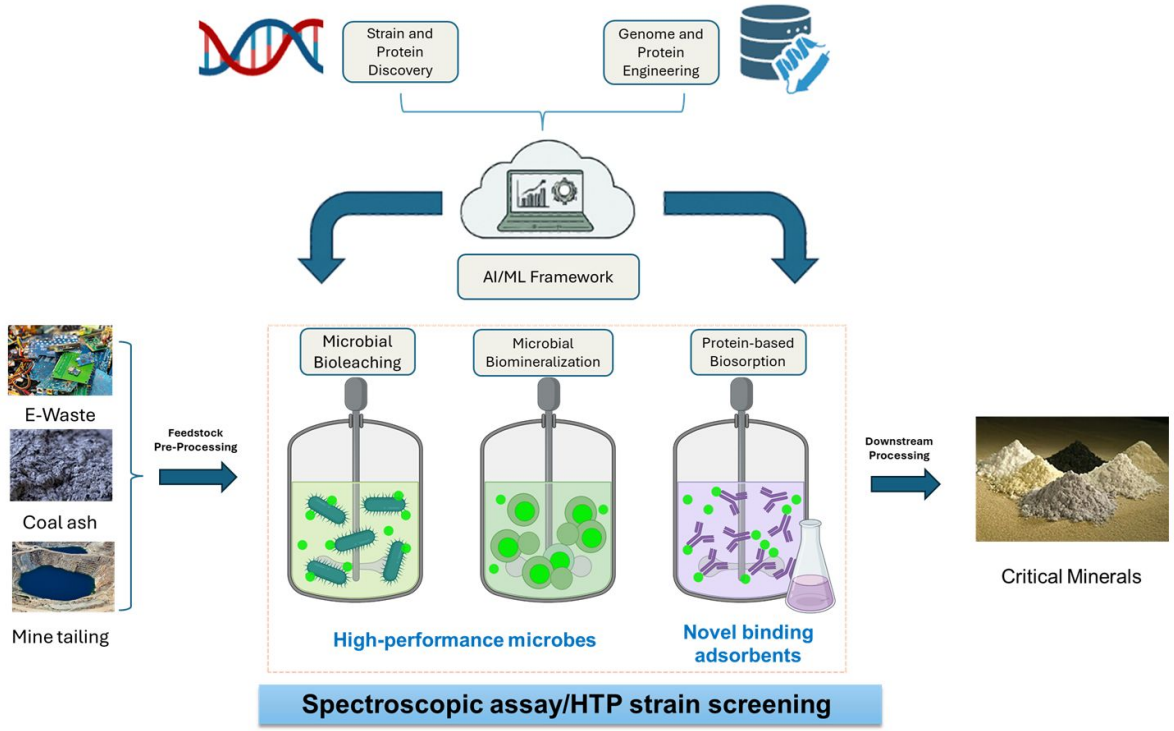
Goal: To understand the fundamental mechanisms of biorecovery and develop new microbial chassis and bioadsorbent harnessing their natural capacity to efficiently recover REEs through a modular, AI/ML-guided technology.

Task 1: Technical tool development including AI/ML framework, spectroscopic REE characterization assay, and high-throughput (HTP) strain screening and validation pipeline

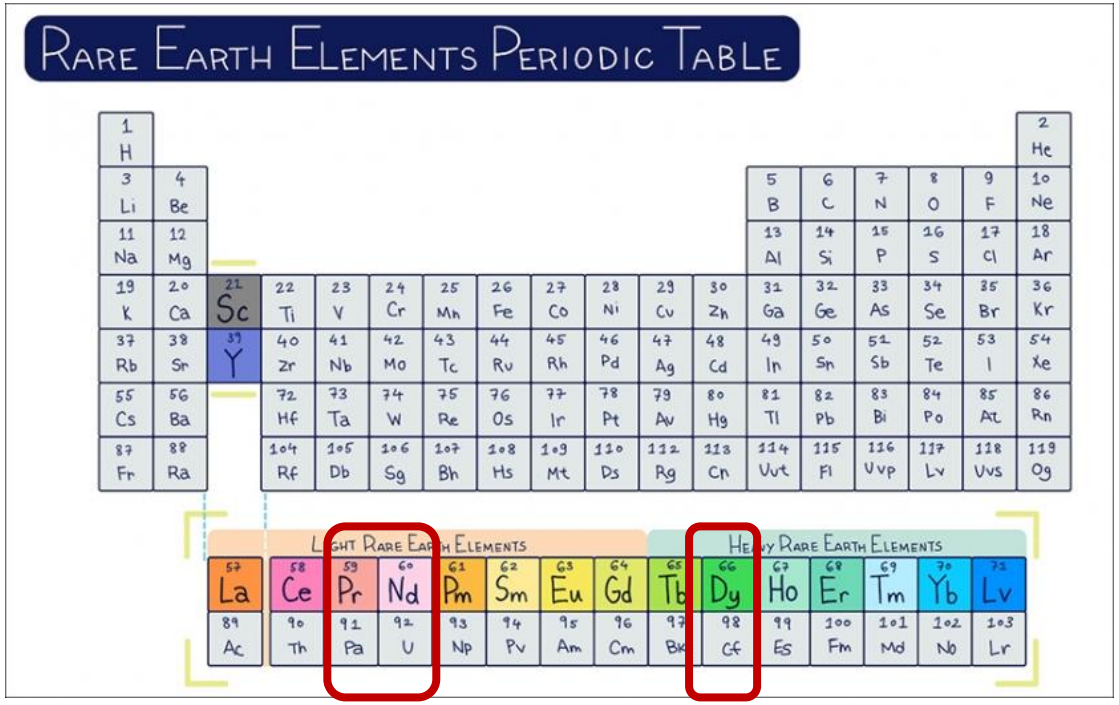
- Task 1.1 Construct Knowledge Graph (KG) and Establish AI/ML framework
- Task 1.2 Spectroscopic assay development
- Task 1.3 Develop HTP strain screening and validation pipeline

Task 2: AI/ML-guided design of microbes for REE accumulation and usage through bioengineering

- Task 2.1 Bacterial strain engineering to improve REE selectivity
- Task 2.2 Expanding fungal chassis for robust biosorption and biomineralization of REEs



Focus on REEs present in E-waste



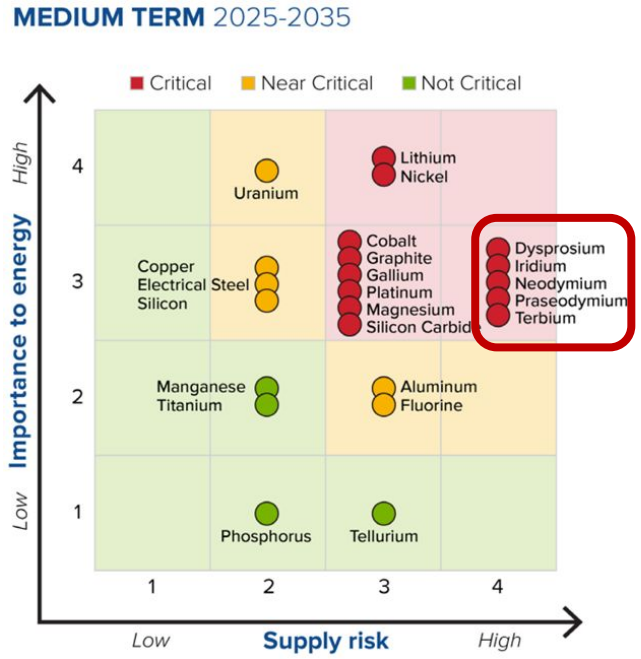


Figure ES.2. Medium-term (2025–2035) criticality matrix

- **Neodymium (Nd):** The most prevalent REE, with concentrations in the magnets as high as 22.9%
- **Praseodymium (Pr)**: Added to magnets to improve properties with concentrations up to 13%
- **Dysprosium (Dy):** Added to magnets to increase thermal stability with concentrations up to 4%

Task 1.1: Construct Knowledge Graph (KG) and establish AI/ML framework

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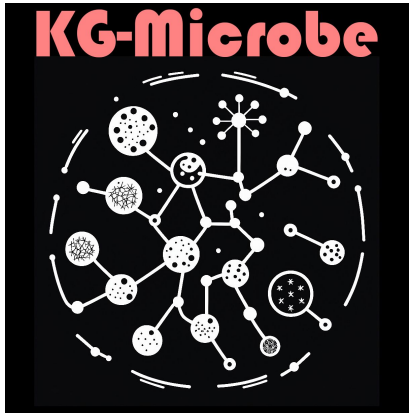

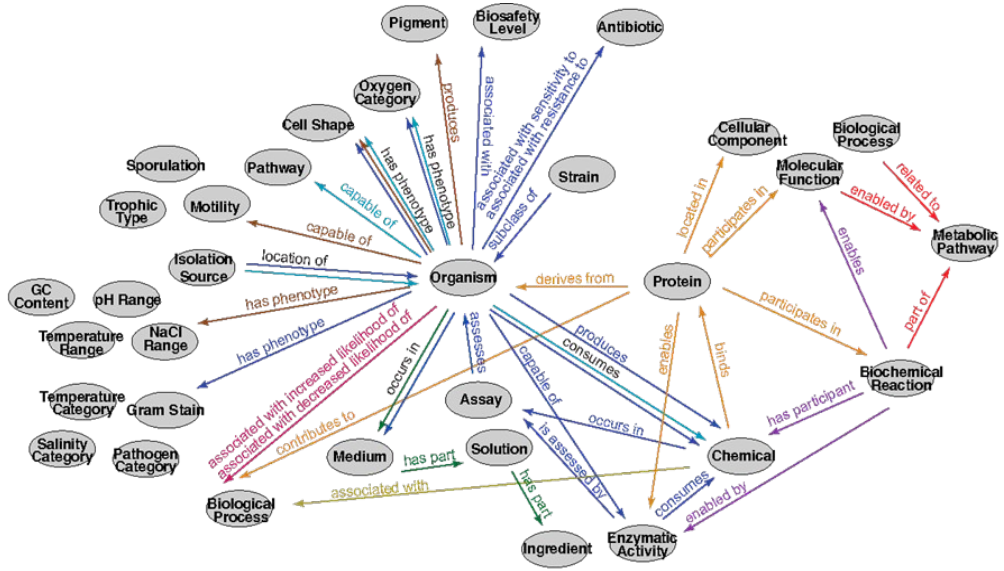

Source	
BactoTraits	Comparative Toxicogenomics Database (CTD)
Bacterial Diversity Metadatabase (BacDive)	MediaDive
UniProt Knowledgebase (UniProt)	UniPathways Ontology (UPA)
Disbiome Database	Rhea
Wallen et al.	Madin et al.

> 3,000 organismal traits
> 30,000 functional/genomic traits

- A KG for microbiology has been developed harmonizing data on organismal, complex traits (such as pH preference) along with genome annotations.
- The data supports AI model training for microbial growth preference prediction.

Santangelo et al., 2025, Caufield et al., 2023, Unni et al., 2022; Joachimiak et al 2021
github.com/Knowledge-Graph-Hub/kg-microbe

Task 1.1 Agentic AI framework for AI-driven CMM experiments

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Task 1.2 Spectroscopic assay
data & analysis

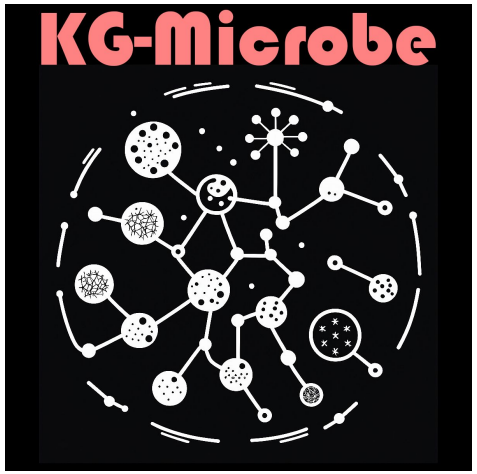
Task 1.3 HTP strain screening ***data & analysis***

Task 2.1 Bacterial strain engineering ***data & analysis***

Task 2.2
Expanding fungal chassis
data & analysis

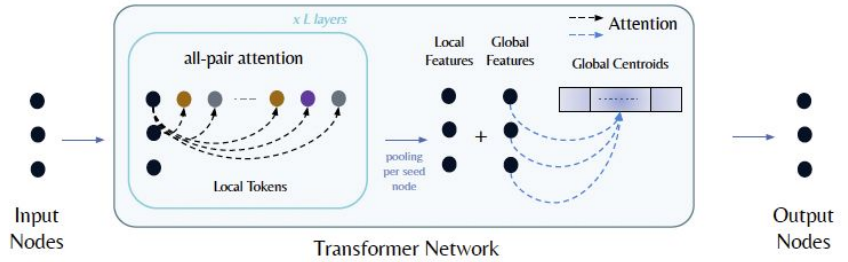
Agentic AI Experiment Orchestrator
Goal → Plan → Execute → Process → Learn

KG-Microbe



Machine-Reading Literature
These isolates grew well with naphthalene as the sole carbon and energy source

AI models



Develop an agentic AI framework using a foundational model KG and graph transformer model to support prediction and generation of content such as experiment design optimizations.

Task 1.2 Spectroscopic assay development to enable HTP detection

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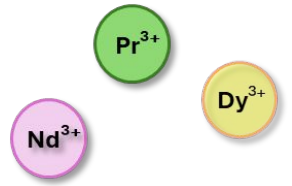
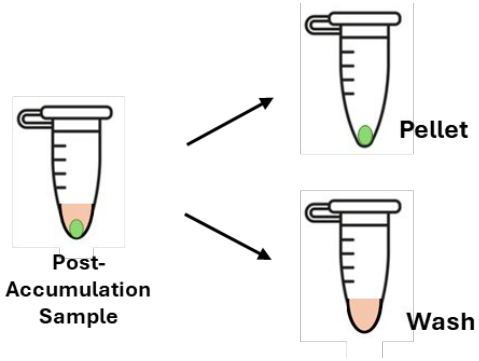
Goal: ICP-OES and HTP Luminescence Assay for REE accumulation



1. Inductively Coupled Plasma – Optical Emission Spectroscopy (ICP-OES)

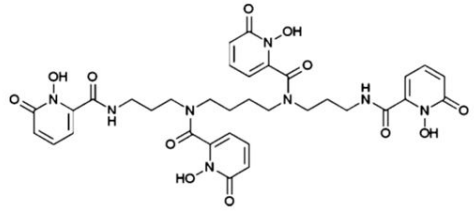
Quantify REEs in digested cell sample and supernatant/wash fractions to assess accumulation of critical materials (Pr^{3+}, Nd^{3+},Dy^{3+}, etc.)



2. Time Resolved Luminescence (TRL) with Microplate Spectrophotometer

Sensitization of Eu^{3+} and Tb^{3+} for TRL quantitation via the Antenna Effect

3,4,3 LI(1,2-HOPO) chelator has high binding affinity towards Ln^{3+} species



3,4,3-LI(1,2-HOPO)

Sturzbecher-Hoehne et al., *3,4,3-li(1,2-HOPO): In vitro formation of highly stable lanthanide complexes translates into efficacious in vivo europium decorporation* **2011**

Pallares et al., *Precision engineering of 2D protein layers as chelating biogenic scaffolds for selective recovery of rare-earth elements* **2022**

Task 1.2 Spectroscopic assay development to enable HTP detection

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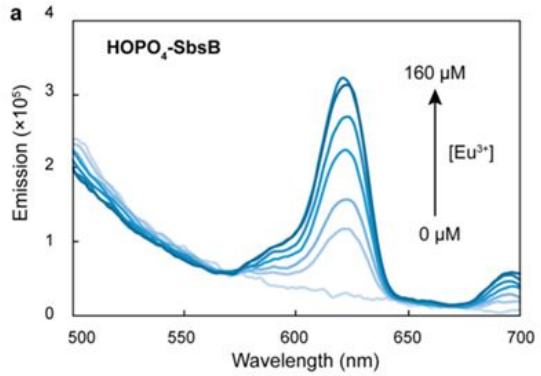
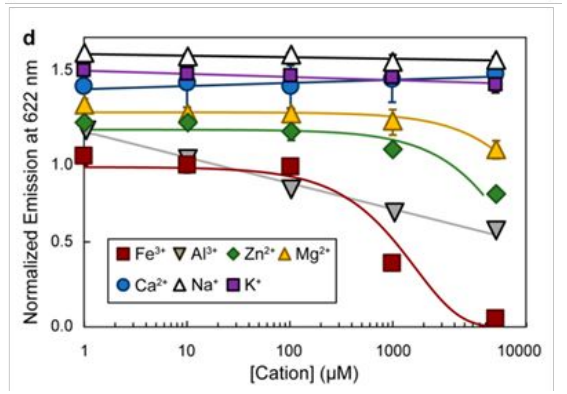
Time Resolved Luminescence (TRL) and Antenna Effect

- TRL enables the quantification of target analytes in cells with high specificity
- Lanthanide elements (Eu, Tb) exhibit distinct luminescent signature that are enhanced through the binding of antenna molecule.
- 3,4,3 LI(1,2-HOPO) binding has demonstrated great effectiveness in enabling lanthanide fluorescence in biological matrices



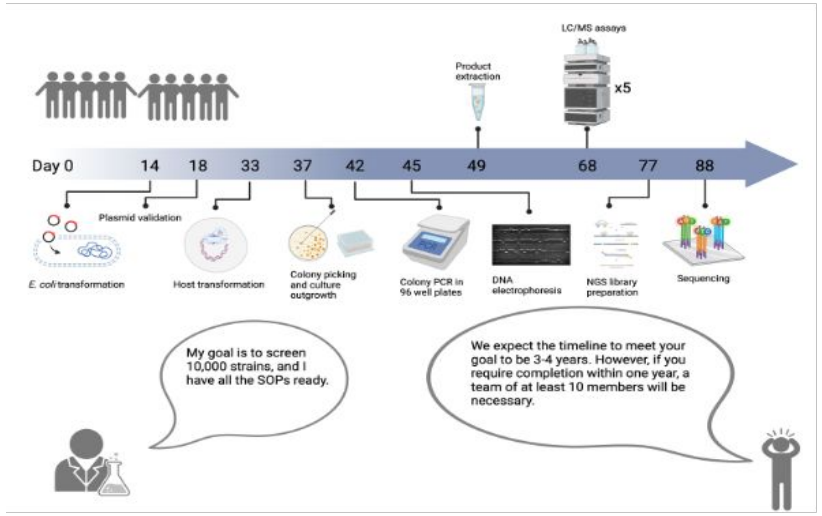


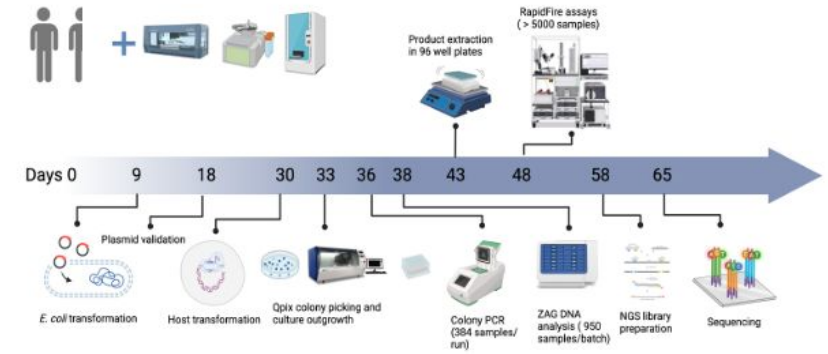
- 3,4,3 LI(1,2-HOPO) and derivatives can be synthesized in house at the gram scale and applied to the HTP workflow.

Previous Research

- 2D biogenic scaffold with HOPO subunits used to capture Eu^{3+}
- Investigation of emission intensity of 3,4,3 LI(1,2-HOPO) lanthanide complexes across the series.

Task 1.3 HTP strain screening and validation pipeline-prior work





Available High-Throughput Process Workflows

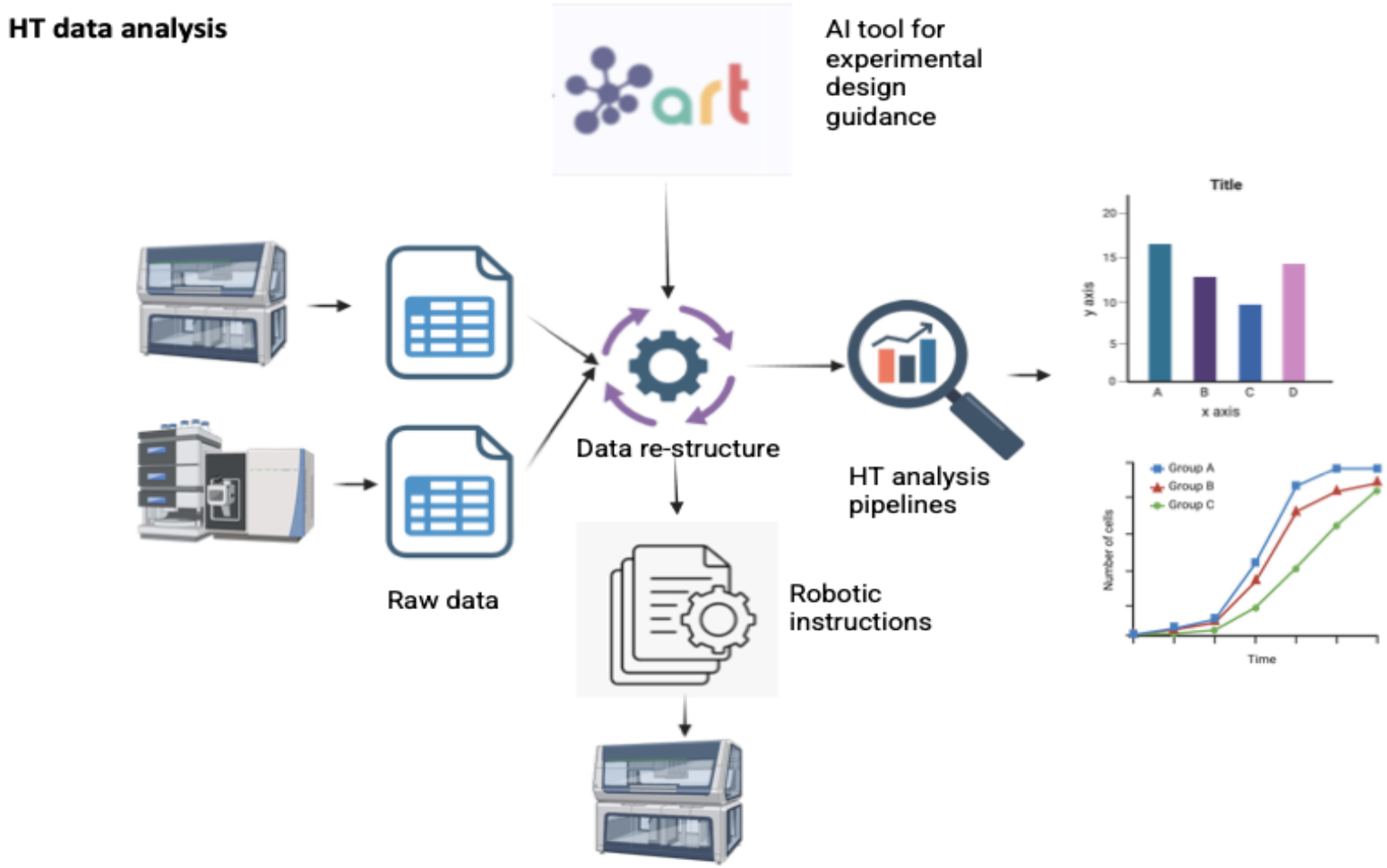
1. **Automated *E. coli* transformation** – 96 different constructs per batch
2. **HT colony validation** – NGS platforms, 1,536 samples per batch
3. **Automated colony picking** – QPix, >1,536 colonies per batch
4. **Media plate preparation** – Standardized and scalable workflow
5. **Glycerol stock & production plate preparation** – 6,144 samples per batch
6. **Strain adaptation process** – 6,144 samples per batch
7. **Sample preparation** – NGS libraries, RapidFire assays, or proteomics (6,144 samples)
8. **Automated fluorescent & OD measurements** – 6,144 samples per batch (5 hours of processing time)
9. **HT variant sequencing** – Applications include RB-Tn, oligo pool, and promoter libraries (1,536 strains per batch)
10. **NGS analysis pipelines** – Variant detection and data processing

Workflows in Development

1. **Streamlined workflows for complex buffer preparation**
2. **HT data analysis pipelines for plate reader datasets**

Kuo et al., 2025. A Modular High-Throughput Pipeline for Automated Microbial Strain Engineering: From Colony Picking to Phenotypic and Genotypic Analysis. doi.org/10.17504/protocols.io.261gerqkdl47/v2

Task 1.3 HTP platform data processing pipelines-prior work



- Data-formatting pipeline, which can:
  a. **Incorporate experimental designs** from AI tool (e.g. ART) and generate robotic instruments for automation
  b. **Re-organize raw data from instruments** (e.g. HPLC, spectrometer) for downstream analysis
- **HTP data analysis pipelines**, which automatically analyze the processed data and generate graphical outputs.
- **NGS variant detection pipelines** that process Illumina sequencing data to identify genetic variants in each strain.

Unpublished analysis pipelines designed and developed by Rita Kuo

Task 1.3 Establish HTP pipelines for REE biorecovery-Approach

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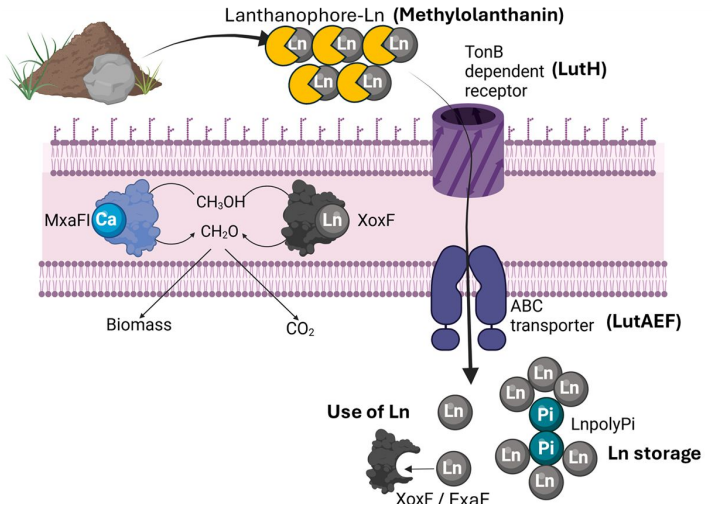
- **Screen growth rates:** SNPs in the regulator correlate with both REE accumulation and growth rates (control: WT strains).
- **Quick assays:** Color-based assays that require pH adjustment; UV-vis; Input from task 1.2
- **HTP genotyping for single strain**
- **ICP–OES**: High-Throughput workflows for ICP sample preparation
- Others
  - Media optimization to improve growth or accumulation of REE in strains
  - Development of tolerance ALE workflow of the strains with REE
  - Leverage protein engineering tools to help with REE intake and capsule formation.

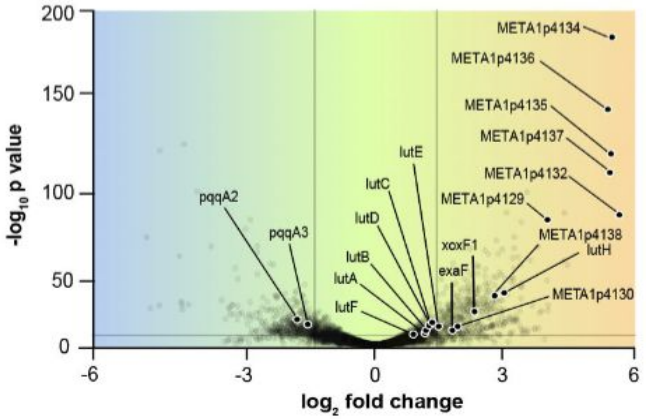
Task 2.1 Bacterial strain engineering to improve REE selectivity

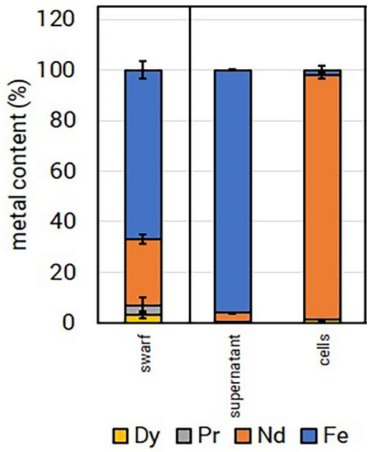
- Martinez-Gomez lab has defined the machinery necessary for microbes to use lanthanides that includes the secretion of lanthanide chelators, specific transport systems, enzymes that use lanthanides as cofactors and a storage process of lanthanides inside the cell.

- They have developed strains that are able to hyperaccumulate lanthanides (specifically Nd, Pr, and Dy) from electronic waste with high selectivity for lanthanides and not iron or Boron.



Berkeley
UNIVERSITY OF CALIFORNIA





Transcriptomic profiles of REE-utilizer strains enables the identification of gene clusters that produce REE-chelators

Task 2.1 Bacterial strain engineering to improve REE selectivity-Approach

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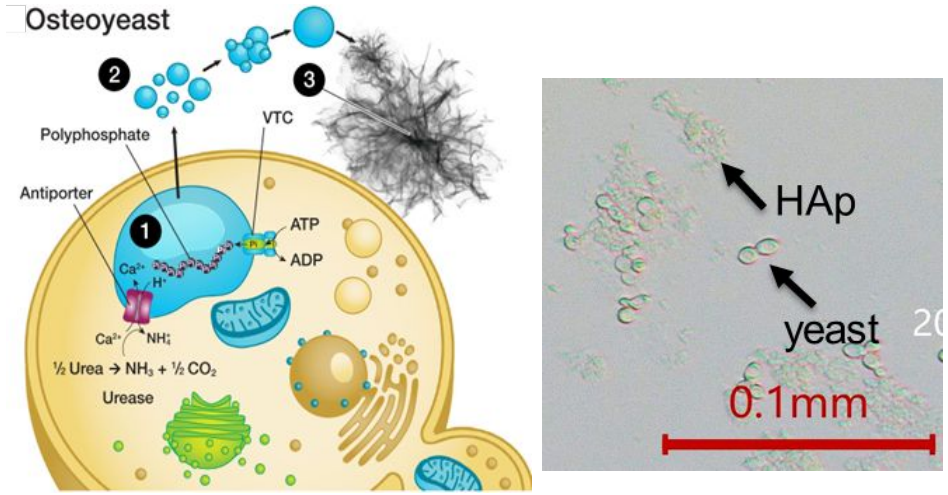
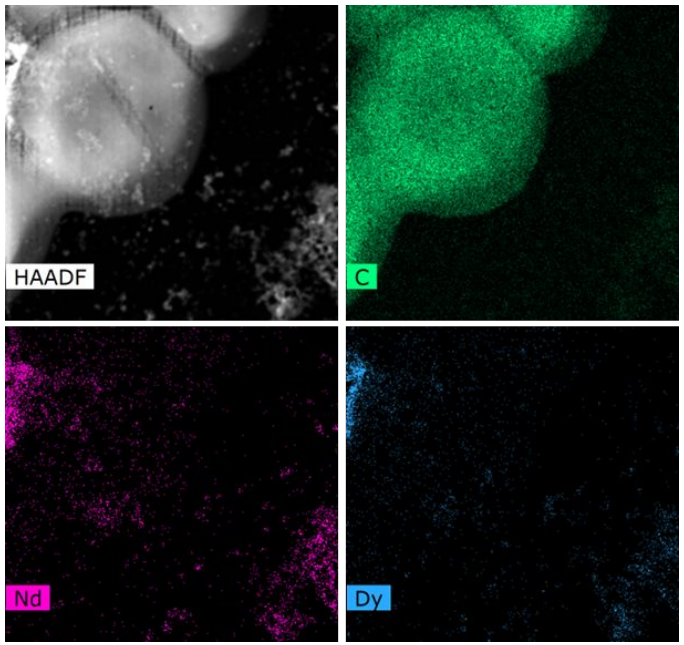
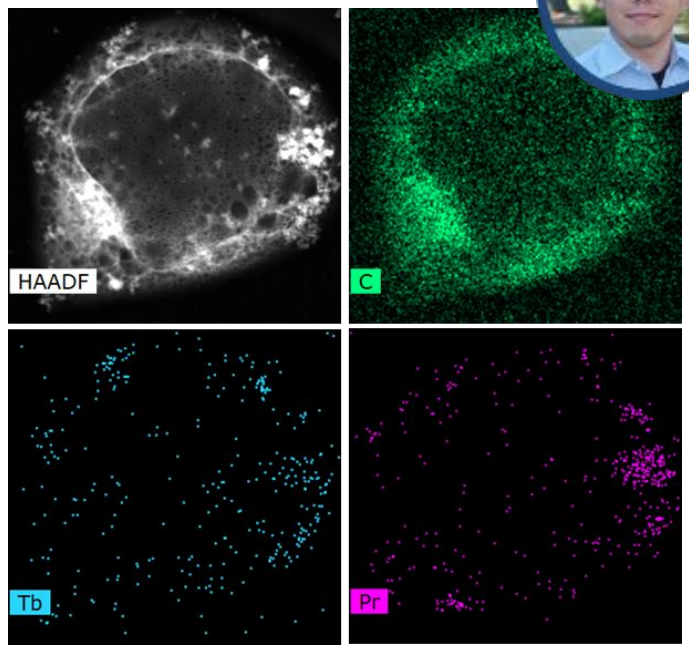
- Engineer bacterial strains to hyper-accumulate with high selectivity for chosen REEs.
- Identify and validate novel biosynthetic clusters producing novel lanthanophores through transcriptomic profiling along with genomic analysis .
- The strains with novel biosynthetic clusters will be screened for growth rate and growth yield using different REEs.

Task 2.2. Expanding bacterial and fungal chassis for robust biosorption and biomineralization-previous results







Engineered yeast enables accumulate Ca^{2+} and form HAp

- REE enrichment occurred at the yeast cell surface, potentially reflecting adsorption or localized precipitation.

Müller et al., *Nat. Commun.*, 2025

Task 2.2. Expanding bacterial and fungal chassis for robust biosorption and biomineralization-approach

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Strains known for bioleaching
- *Acidithiobacillus ferrooxidans*
- *Acidithiobacillus thiooxidans*

Strains known for biosorption
- *Methylobacterium extorquens*

Strains identified through HTP screening (Task 1.3)

Universal strain engineering tool
Wang et al., *Nature Microbiology,* 2019

Characterization of molecular mechanisms underlying REE recoveries

Enhancing bioleaching, biosorption, biomineralization activity

Functional activation and discovery of novel metallophores

- Apply Chassis-independent Recombinase-Assisted Genome Engineering (CRAGE) to domesticate and optimize both well-characterized extremophiles and newly discovered bacterial isolates for high-efficiency biomineralization of REEs
- Engineer acid-, salt-, and temperature-tolerant fungal chassis, leveraging prior success in biomineralization

Expected Outcomes: Bioprocessing platform enables REE circular economy

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We will validate key biological design rules and identify at least one novel microbial chassis, along with previously uncharacterized components of the lanthanome, to enable efficient REE recovery from E-waste. Major deliverables include:

- Establish an AI/ML-guided framework to support core task activities and serve as a unified source of data integration and model predictions
- Establish spectroscopic assays for REE quantification in a high-throughput manner
- Establish HTP pipeline to screen and validate high performance microbial strain/consortia for REE bioaccumulation
- Identify and validate at least one microbial strain with high REE bioaccumulation through AI/ML