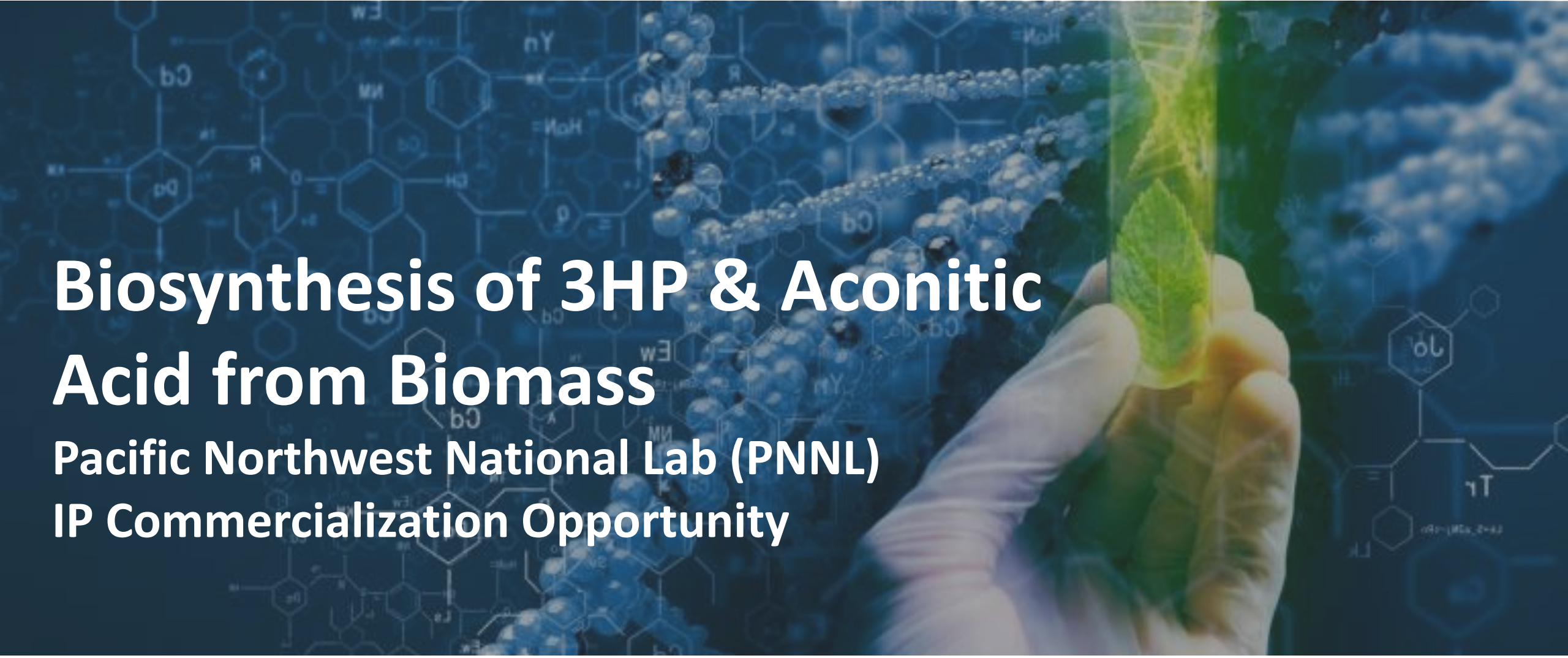


Biosynthesis of 3HP & Aconitic Acid from Biomass

Pacific Northwest National Lab (PNNL)

IP Commercialization Opportunity

tradespace

Pacific Northwest
NATIONAL LABORATORY

Pacific Northwest National Lab (PNNL) Licensing & Commercialization Opportunity

Pacific Northwest NATIONAL LABORATORY

Opportunity: PNNL has engaged Tradespace to approach select partners to commercialize biosynthetic production processes through co-development and IP licensing

Technology: PNNL has genetically modified fungi to produce 3HP or aconitic acid – key inputs for biodegradable plastics and biocompatible materials– using biomass instead of fossil fuels

Key Benefits of Aconitic Acid and 3HP Biosynthesis Method:

- Produces biodegradable & biocompatible end-products
- Useful as plasticizer for polymers, biomaterial, or natural flavoring
- No fossil fuels used in production
- Higher titer than other production methods

Opportunity Snapshot:

Available IP

- One Granted Patent (US10947548B2)
- Two Patent Applications
- Expertise & Data Package
- Access to Research Team

Maturity Level

- Synthesized in lab-scale reactor (10L)

Potential Partnership Models

Patent License | Collaborative R&D | Sponsored Research

tradespace

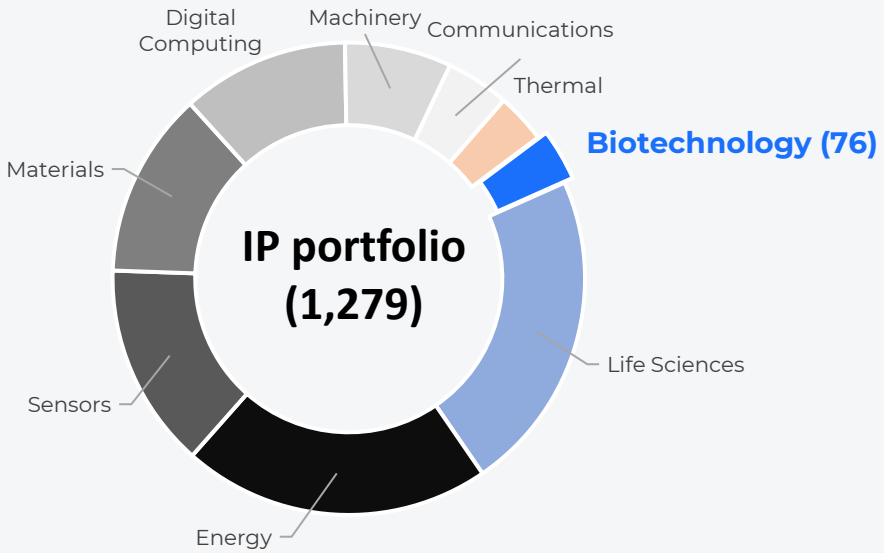
Pacific Northwest National Lab Snapshot

Pacific Northwest NATIONAL LABORATORY

Leading Department of Energy lab with focuses in chemistry, data analytics, life science, and technological innovation in energy resiliency and national security. Based in Richland, WA

Annual R&D Spend: $1.01B **Staff:** 4,722

PNNL Biosynthesis Expertise



- Deep expertise in fungal genetics
- Research staff includes leading expert in multiomics and proteomics
- Lead testing lab for the Agile Biofoundry – a DOE-wide biomanufacturing effort

Agile BioFoundry

National Labs with Biosynthesis IP		
Rank	Organization	IP Strength
#1	Lawrence Livermore	75%
#2	Sandia National Lab	74%
#3	**PNNL**	**72%**
#4	Oak Ridge National Lab	70%
#5	Idaho National Lab	63%
#6	Naval Research Lab	62%
#7	Army Research Lab	60%
#8	Argonne National Lab	58%
#9	NASA	52%
#10	Los Alamos National Lab	51%
#11	Brookhaven National Lab	49%
#12	Air Force Research Lab	38%
#13	Savannah River Nat'l Lab	36%

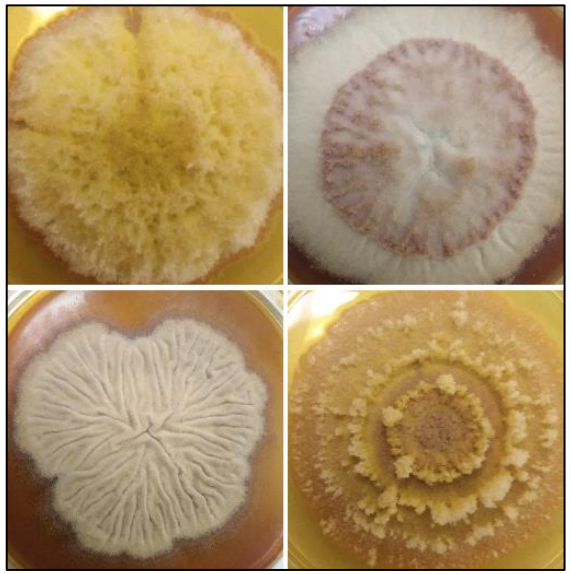
Why Use Fungi for Chemical Biosynthesis?

Pacific Northwest NATIONAL LABORATORY

Biosynthesis Overview: Multi-step, enzyme-driven process for converting low-value feedstock into valuable chemicals using living organisms like fungi, bacteria, or yeast

Benefits

- **Low Cost:** Can leverage low-cost, readily available feedstocks like biomass (Lignocellulosic)
- **High Titer:** Can result in significantly higher yields than bacteria- or yeast-based approaches
- **Existing Infrastructure:** Fungi-based biosynthesis is already widely used for other chemical synthesis processes
- **Simpler Fermentation Process:** Can tolerate low pH; free-acid is easier for separation



Aspergillus fungi used for biosynthetic production of 3HP & Aconitic Acid

tradespace

Use-Cases for Monomers Produced by PNNL's Biosynthesis Process

Pacific Northwest NATIONAL LABORATORY

Monomer Produced by Biosynthesis	Intermediates	End-Uses
3-hydroxypropionic acid (3HP)	Acrylic Acid, Methyl Acrylate, Acrylamide, Propenediol	• Biodegradable polymers / plastics • Hygienic medical products • Water treatment chemicals • Skin care (Propendiol)
Aconitic Acid	Pentanediol, Itaconic Derivatives, Pyrrolidones, Esters	• Biodegradable polymers / plastics • Biocompatible material for implants, drug delivery, & tissue engineering • Flavoring agent

tradespace

Technology Overview: Biosynthesis of 3HP and Aconitic Acid using Aspergillus Fungi

Pacific Northwest NATIONAL LABORATORY

IP Snapshot

Patents: 1 Granted Patent; 1 Application; 1 Divisional Application

Geography: US

Research Team Available: Yes

Technical Data Package: Yes

IP Availability: Patent License; Collaborative R&D; Sponsored Research

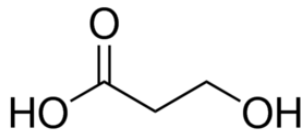
Technology Maturity Level

- *Synthesized in lab-scale (10L) bioreactor*

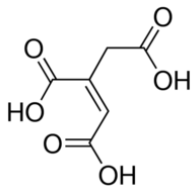
Technology Description

US10947548B2

Genetically-modified, recombinant *Aspergillus pseudoterreus* fungi capable of producing aconitic acid or 3-hydroxypropionic acid (3HP) at high concentrations and high selectivities from low-cost biomass feedstocks.



3-hydroxypropionic acid (3HP)



Aconitic Acid

Key Technical Features & Benefits

- **High-Titer (Concentration):**
  - **3HP:** 40g/L (3X increase from unmodified Aspergillus fungi)
  - **Aconitic Acid:** 50g/L
- **Environmentally-Friendly:** Biodegradable (& biocompatible) alternative to producing polymers with fossil fuels
- **Low pH:** Does not require additives to maintain neutral pH (which would increase cost)
- **Industrially-Relevant Fungi:** *Aspergillus spp*. Already used for producing small molecules and enzymes in large bioreactors (e.g., 100,000L airlift reactors)
- **AI / ML:** Tools for genetic modification use ML models that significantly reduce time for further genetic modification of aspergillus or other fungi

tradespace

Engagement Opportunities

Tradespace has been engaged to facilitate industry collaboration and technology transfer for PNNL's IP covering biosynthesis of 3HP and Aconitic Acid using *aspergillus*

Potential collaboration models include:

- Patent License
- Cooperative Research and Development Agreements (CRADAs)
- Sponsored Research
- Collaborative Research

Please contact Alec Sorensen, CEO of Tradespace, for further information regarding the opportunity

Contact:
Alec Sorensen
CEO Tradespace
alec@tradespace.io
804-836-7938

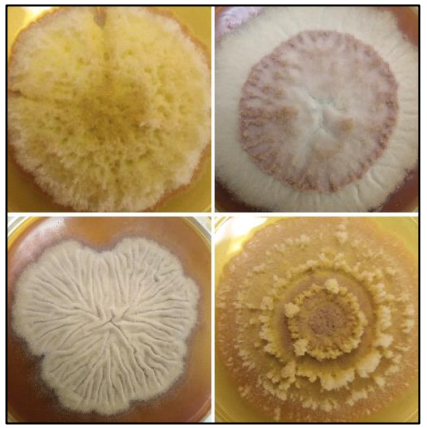
tradespace

Appendix: Overview of *aspergillus* Biosynthesis Process

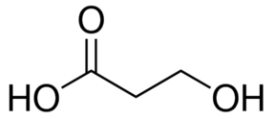
Fungi Overview:
Aspergillus pseudoterreus & Aspergillus niger

Pacific Northwest
NATIONAL LABORATORY

- **Industrially Relevant:** Used for producing commodity fuels, chemicals, and enzymes in large bioreactors
  - **e.g.,** citric acid in 100,000L airlift reactors, ~3M ton market
- **Genetically tractable:** Genome sequenced, genome scale metabolic model developed
- **High flux through glycolysis toward TCA cycle derived organic acids**
  - **E.g.,** A. pseudoterreus ATCC 32359 makes 60 g/L itaconic acid
- **Grows and produces at pH 1-3, hence it produces acids, not salts**
  - **Separations:** High titer, free acid, crystallization possible
  - Does not require lime to maintain neutral pH, sulfuric acid for post process acidification, which generates gypsum waste



tradespace

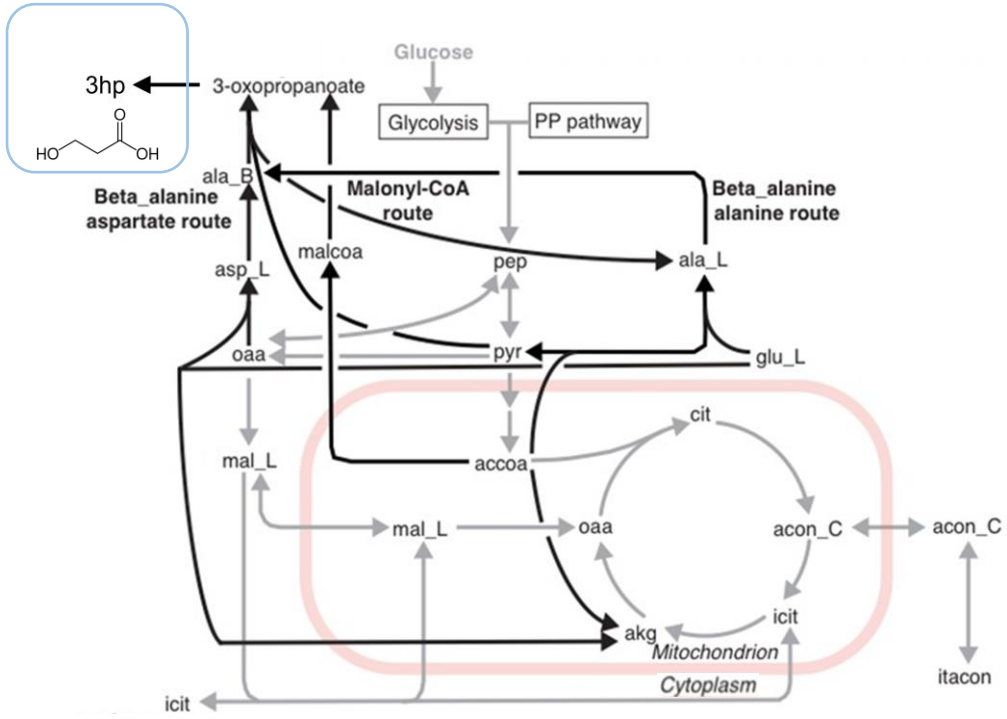


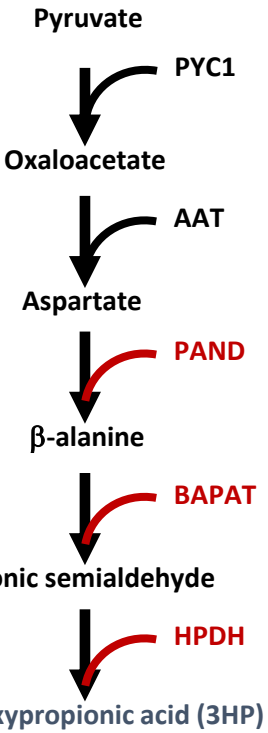
Biosynthesis Product #1:
3-hydroxypropionic acid (3HP)

Pacific Northwest NATIONAL LABORATORY

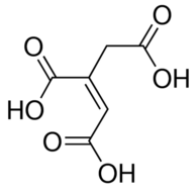
- **Intermediate to acrylic acid & acrylonitrile**
- **Heterologous pathway (prokaryotic)**
- **Beachheads:** Pyruvate, oxaloacetate
- **PNNL has complimentary portfolio of acrylonitrile IP**

Biosynthetic pathway for 3HP production from biomass feedstock





tradespace

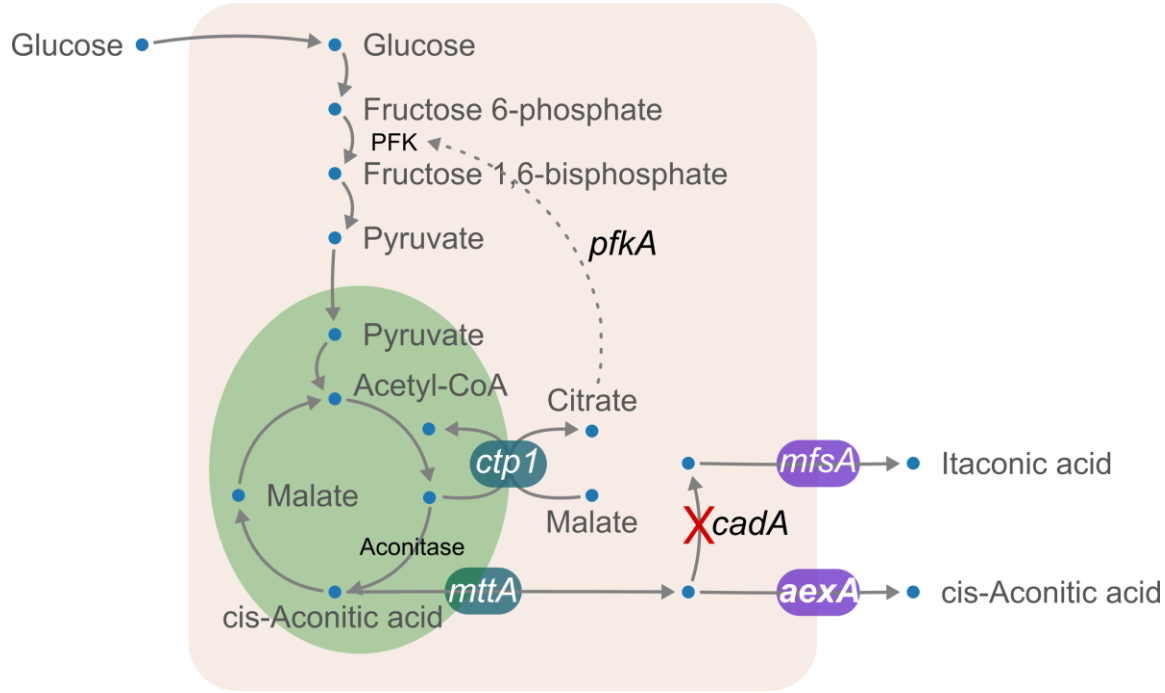


Biosynthesis Product #2:
Aconitic Acid

Pacific Northwest
NATIONAL LABORATORY

- A 6-carbon tricarboxylic acid, like citric acid
- Beverage acidulant, industrial chelator/modifier (cement) etc.
- Central metabolite with transport limitations
- **Beachheads:** Pyruvate, oxaloacetate, acetyl-CoA

Biosynthetic pathway for aconitic acid production from biomass feedstock



tradespace