



# The Biotransformation of All Chemistry

Leveraging the **Fastest** Engineering Technology  
with the **Deepest** Enzyme Database



Our mission is to  
create **robust biosolutions**  
for a **greater tomorrow.**







## **Peyman Salehian**

### **Co-Founder, CEO**

- PhD in Chemical & Molecular Engineering (NUS)
- Building and scaling biotech processes
- Serial entrepreneur



## **Akbar Vahidi**

### **Co-Founder, CTO**

- PhD in Chemical & Molecular Engineering (NUS)
- Inventor of Allozymes' technology
- 10 years experience in enzyme engineering



**allozymes**





# Our

# Team

- 35+ Employees
- 100+ Publications
- 95 Years of Industry experience in Protein engineering, Biology, Microfluidics, Sequencing, and Computing

## Business



**Audrey Robic, PhD, Pharm D**  
**Director of Business Development**  
15 years of experience with enzyme engineering business development



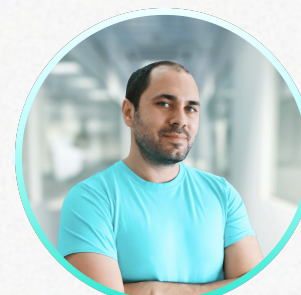
**Anushree, PhD**  
**Business Development Manager**  
7 years of experience with biopharma business development

## Microfluidics



**Patrick Tan, PhD**  
**Senior Microfluidics Scientist**  
14 years of experience in microfluidics in addition to lecturing

## Data



**Tiago Resende, PhD**  
**Senior Computational Biologist**  
9 years of experience in computational biology

## Bioprocessing



**Sanjay Dsouza**  
**Senior Downstream Process Scientist**  
15 years of experience in downstream process engineering



**Prasanth Baku**  
**Senior Bioprocess Engineer**  
9 years of experience in downstream process engineering

## Bioengineering



**Pradeep Nair, PhD**  
**Lead Protein Scientist**  
14 years of experience in protein biophysics



**Shelly Cheng, PhD**  
**Senior Protein Scientist**  
10 years of experience in protein engineering



**Balaji Sekar, PhD**  
**Strain Engineering Scientist**  
10 years of experience in strain engineering





# From humble experiments to the world's **fastest** enzyme engineering powerhouse



Pre-2020

2020

2021

2022

2023

2024

## University Program

## Microfluidics Technology Patented

## \$5 Million Seed Round Secured

## Successfully Completed 1st Customer Project Worth \$500,000

## \$15 Million Series A Round Secured

## 3 Major Biosolutions Launched + Enzyme Data Infrastructure Completed

First 3 years of technology development funded by Singapore's Pharma Innovation Program.

- Company established
- Pre-seed funding secured
- Relocated to shared lab space
- Hired 3 employees

- Achieved 10x improvement in Microfluidics Technology
- Relocated to 2000 sq ft private lab
- Expanded team to 10 employees

- Achieved 30x improvement in Microfluidics Technology
- Launched screening service with 2 clients
- Expanded team to 17 employees

- Achieved fastest Microfluidics Technology worldwide
- Secured 3 new service customers
- 2 biosolutions for chemical & personal care and 1 for the food industry announced
- Moved to 10,000 sq ft lab
- Expanded team to 30 employees

- Automated Microfluidics post-sorting for data collection
- Launched Biosolution for the food industry
- Launched 2 Biosolutions for the Chemical & Personal Care industries
- Opening of European Office
- Aiming for 4 new screening service customers





## Insight into the Current Technology

Conventional enzyme engineering impedes our biotransformation goals

### Low Success Rate

**10–20%**

- Small library
- Small dataset
- No robust modelling
- Reduced sequence space covered

### Slow

**1–2 Years**

- An average 10 rounds of engineering

### Expensive

**USD  
1 Million +**

- A significant amount of consumable reagents

## Our Technology is Efficient and Economical

Allozymes has built the leading platform for enzyme engineering across all industries.

### High Success Rate

**90-100%**

**Up to 20 Million  
screenings per day**

- High sequence space covered
- Data captured systematically
- Heavily trained Machine Learning

### Fast

**2–6  
Months**

- Faster enzyme variants screening
- Minimal Iterations (up to 4 rounds)

### Affordable

**Maximum Value,  
Minimum Cost**

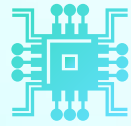
- Higher library screenings for stronger success rate of finding the right enzymes
- Less consumable reagents





# We Deliver Unique Enzyme Sequences Using The Fastest Enzyme Technology Powered By Microfluidics

## Unique Technical Novelties by Allozymes



### Proprietary Chip Designs

Various chip designs for integrated or off-chip droplet incubation



### Generic Detection Assays

Covering several classes of enzymes with generic and sensitive assays.



### Embedded Sorter

Proprietary embedded sorting unit for droplet sorting.



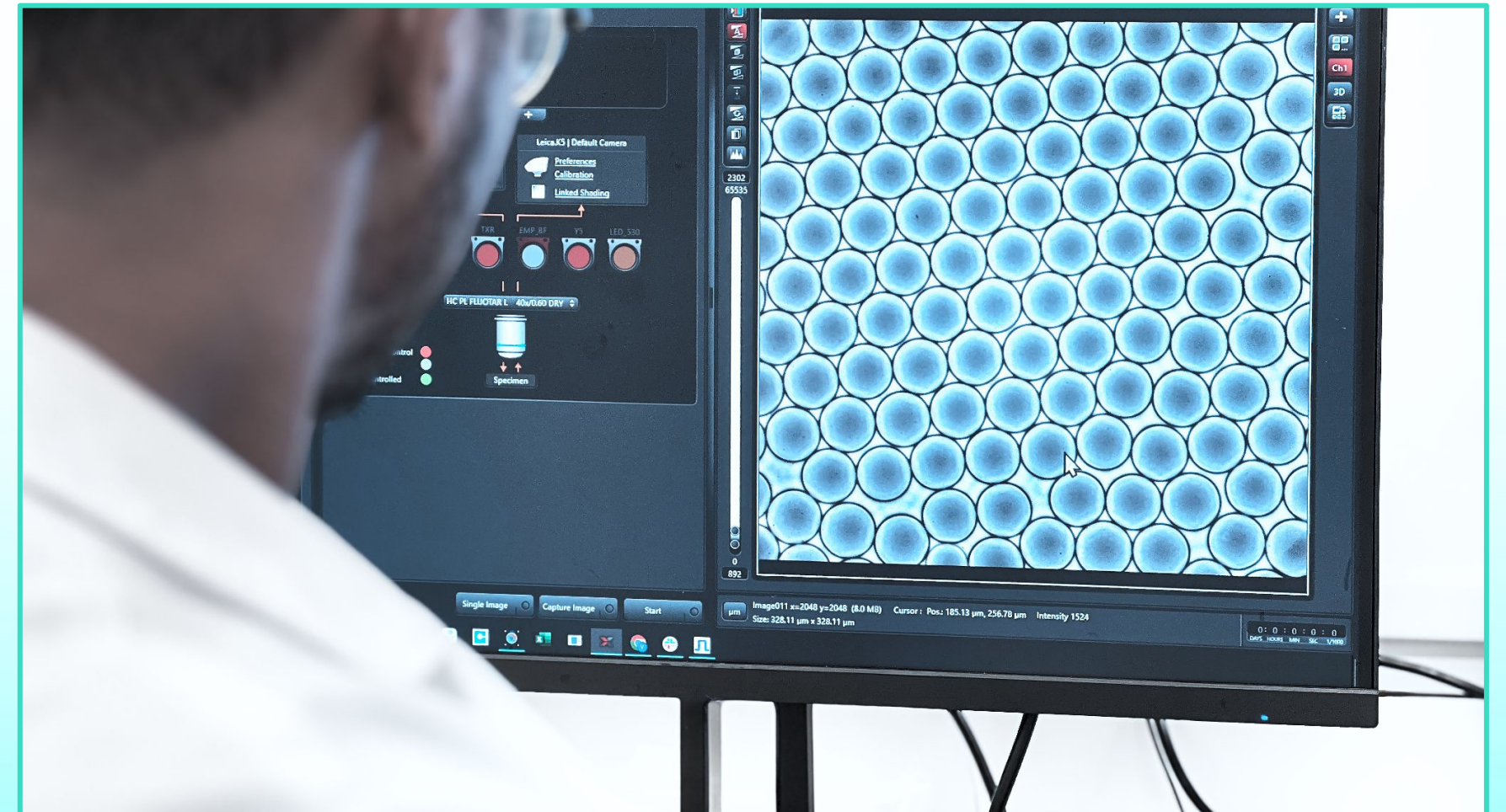
### Post-Sorting Automation

Automatically label and collected the sorted droplets for further analysis



## Directed Evolution with Microfluidics

Allozymes' Microfluidic technology can **miniaturize and automate** enzymatic reactions, allowing for **rapid screening of large libraries** of enzymes against diverse substrates under various conditions.



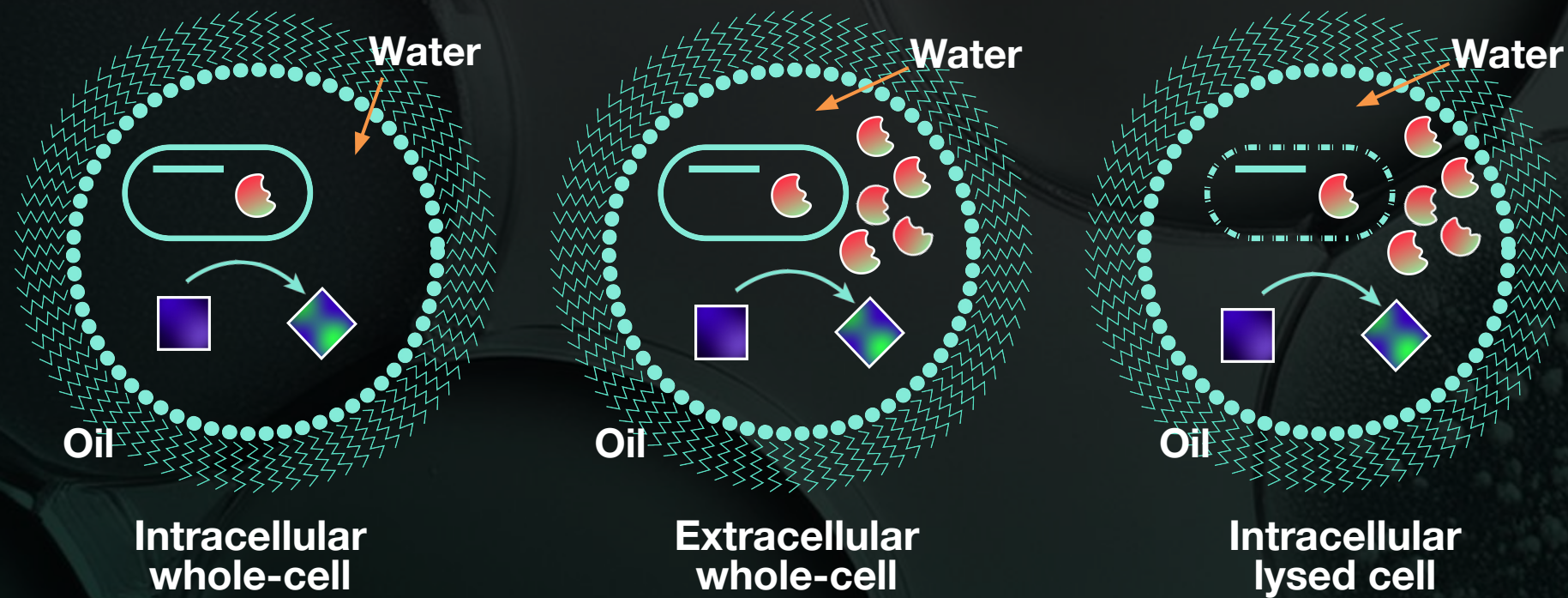




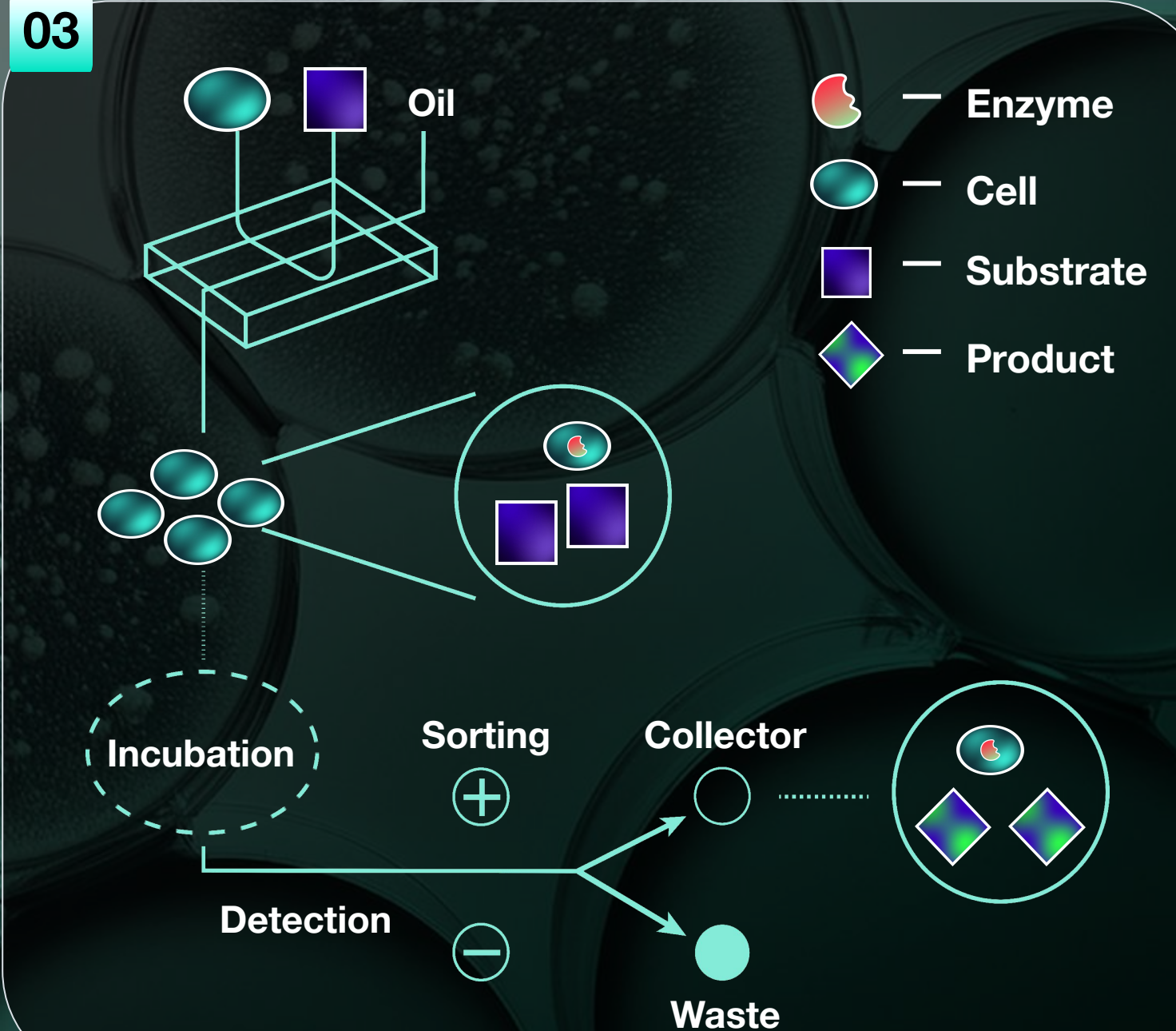
# Versatile Protein Engineering Platform for Diverse Chemistry

**01** We cover several classes of enzymes with versatile and sensitive assays including **Transaminases, Amine Reductases, Amine Dehydrogenases, Hydrolases, Oxidases, Reductases, Isomerases, Transferases, Lyases**

**02** Support cell-free or cell-based systems



- Screening of label-free substrates
- Screening at wide temperature (up to 90C), pH ranges, with miscible solvents
- Detecting enzymes with low to high activity







Protein from Proprietary & Public DataBase

***In-silico***  
**Design**

D

Bioprocess Development  
up to 150 L int.  
CDMO for larger scale

***In-silico***  
**Analyses**

L

# Protein Evolution Workflow

B

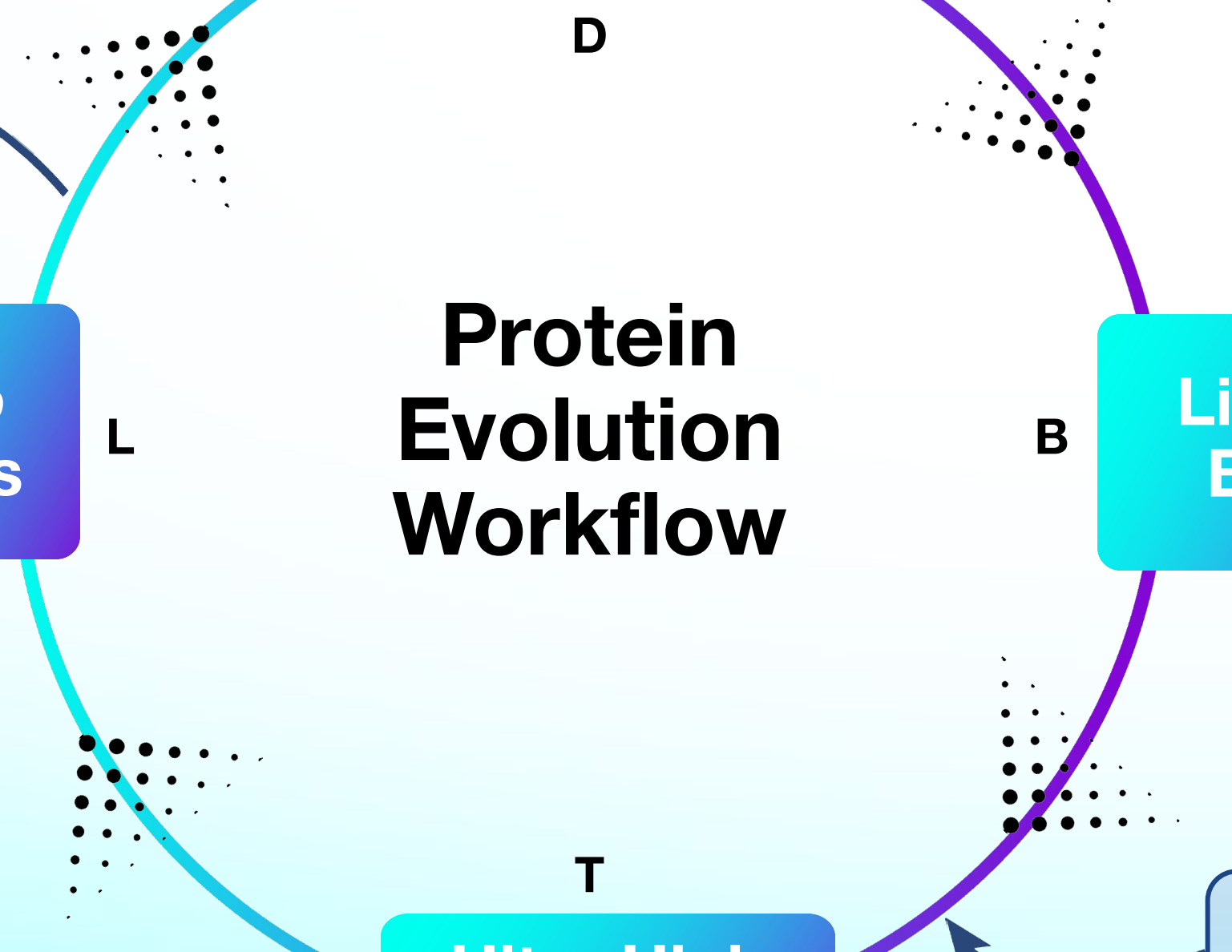
**Library**  
**Build**

T

**Ultra High**  
**Throughput**  
**Screening**

Assay  
Development

**Note:**  
Various Expression Hosts:  
*Pichia, E.coli & Bacillus*







# Allozymes' Services – Enzyme Engineering

## Enzyme Engineering

- Activity
- Stability (pH, thermo, miscible solvent)
- Selectivity: regio and stereoselectivity

## Library Design

- Zero shot & Machine Learning Directed Evolution
- Single-site & combinatorial library
- Molecular docking (binding)

## Enzyme Discovery

- Sequence search (similarity, diversity)
- 3D prediction and crystal structure analysis

## UHT Screening

- Bacteria: E. coli, Bacillus, Yeasts: Pichia, Saccharomyces
- Various Enzyme classes: Transaminases, Amine Reductases, Amine Dehydrogenases, Hydrolases, Oxidases, Reductases, Isomerases, Transferases, Lyases
- Various expression systems (Whole-cell, Lysed-cell, Extracellular)

## Sequencing

- DNA sequencing for various sample types
- Long-read & Short-read
- Whole Genome Sequencing

## Bioprocess Development

- Protein expression and sample
- **USP** – Fermentation optimization
- **USP** – Scaleup (up to 150 L bioreactor internally)
- **USP** – Scaleup >150L with partners
- **DSP** – Purification / Separation & QA-QC





# Allozymes' Services — Strain Engineering

## Strain Engineering

- Pathway assembly and optimization
- Metabolic Engineering
- Strain optimization for target product or protein titer improvements

## Library Design

- Automated virtual assembly
- Genome-scale metabolic modeling
- Genome annotation and primer design
- Genome wide mutant libraries
- Biosynthesis & Retrosynthesis simulations

## Strain Build & Optimisation

- Pathway assembly and optimization
- Strain performance: Titer, yield, rate optimization

## UHT Screening

- Various Strains: E. coli & S. cerevisiae
- Targeted or atheoretical library screenings
- Developing assays and screening flows
- GMO and Non-GMO library screening

## Sequencing

- DNA sequencing for various sample types
- Long-read & Short-read
- Whole Genome Sequencing

## Bioprocess Development

- USP — Fermentation optimization
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# Our Dynamic Business Models

## Fee-for-Service

Support R&D

### Service with Enzyme Ownership

Short-term, one-off R&D project to improve enzyme(s) - Ownership of top variants or full library

### Platform Play

Serve as External R&D partner for multiple R&D projects — Dedicated Team

### Enzyme Manufacturing

Flexibility around bioprocess development and enzyme manufacturing

## Product

Partner Innovative Companies

### Development with IP Licensing

Support partner's discovery and commercial development programs for enzymes & biomolecules

### Product Play

Access Allozymes' specialty product portfolio for target markets & sectors with exclusivity





# Success Stories

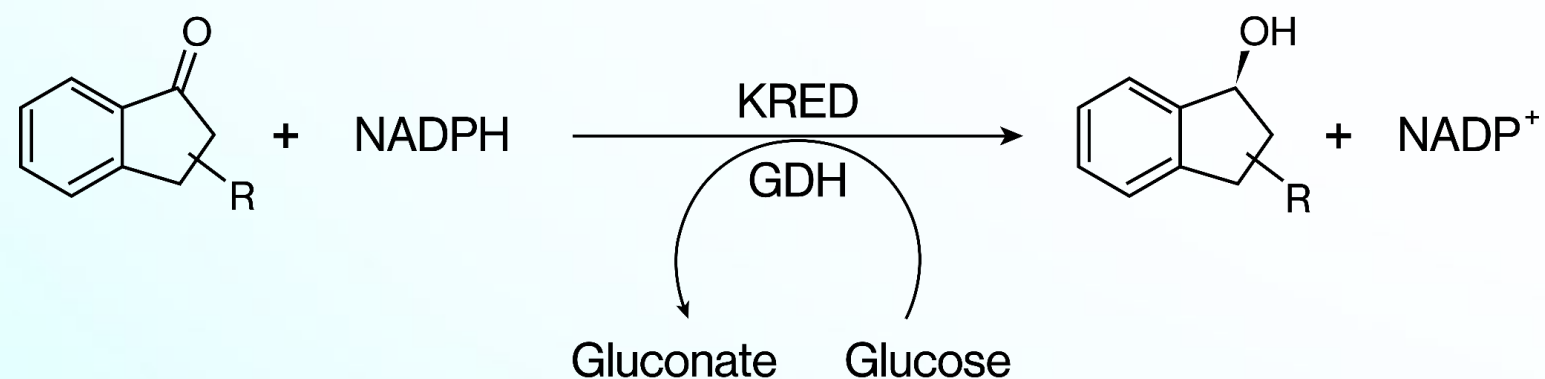




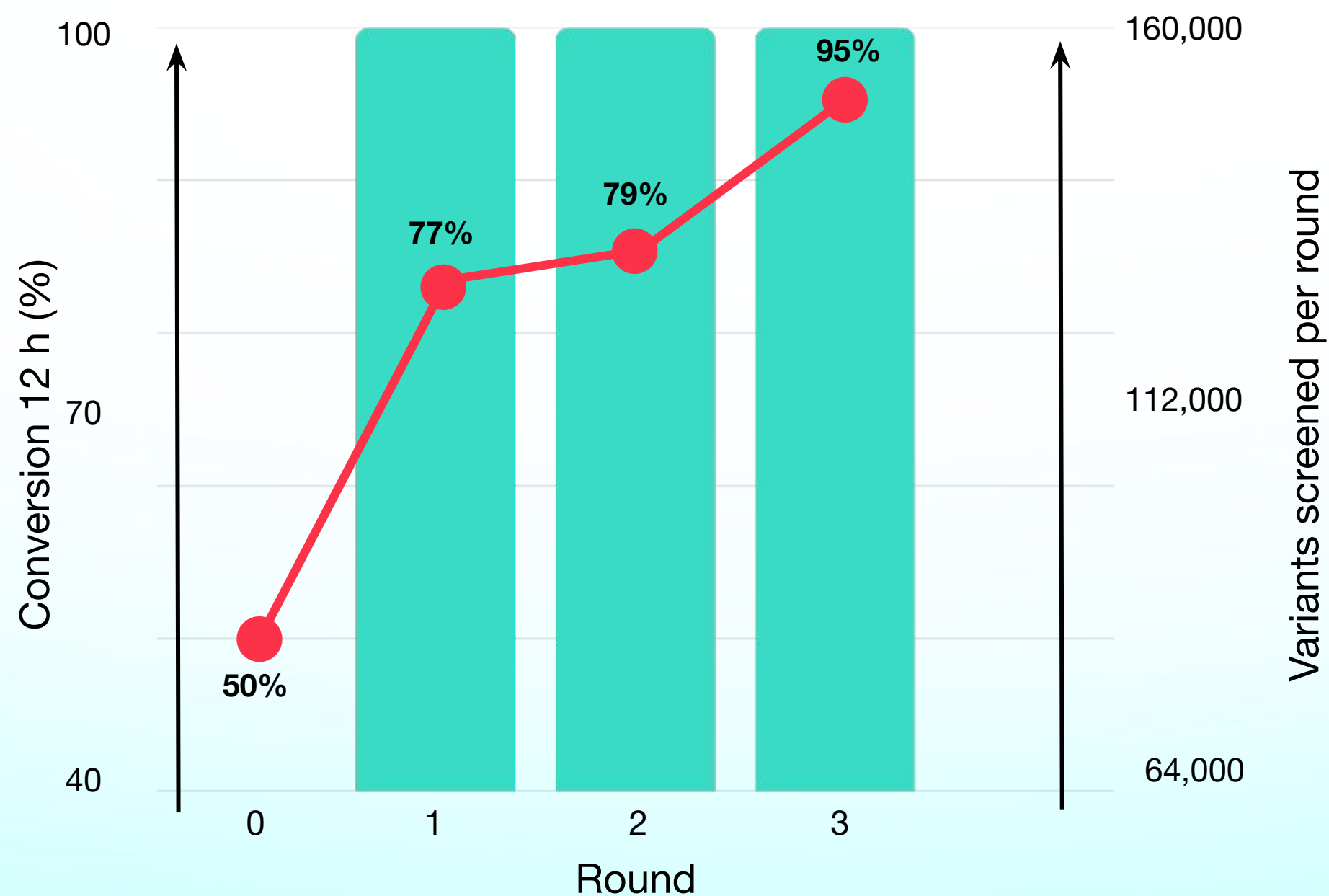


# Enzyme Selectivity – Improved Enantioselectivity of KRED

**Case outline:** Enzyme selectivity is critical for producing chiral alcohol enzymatically



- In 3 rounds, we screened 0.5 million variants
- The new selective enzyme increased conversion from 52% to 95%, with >99% ee
- NADH/GDH/Glucose

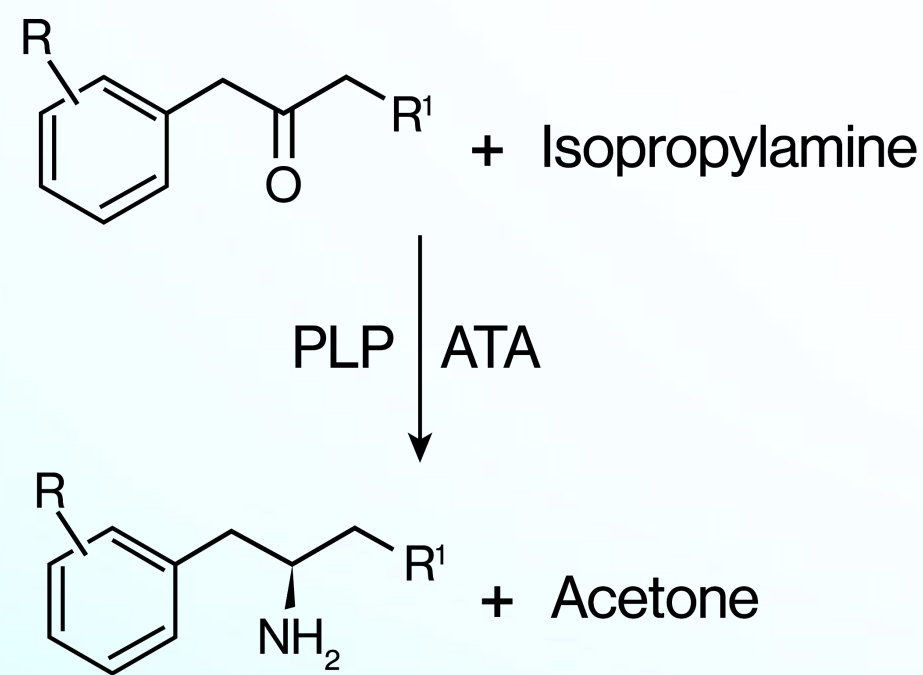




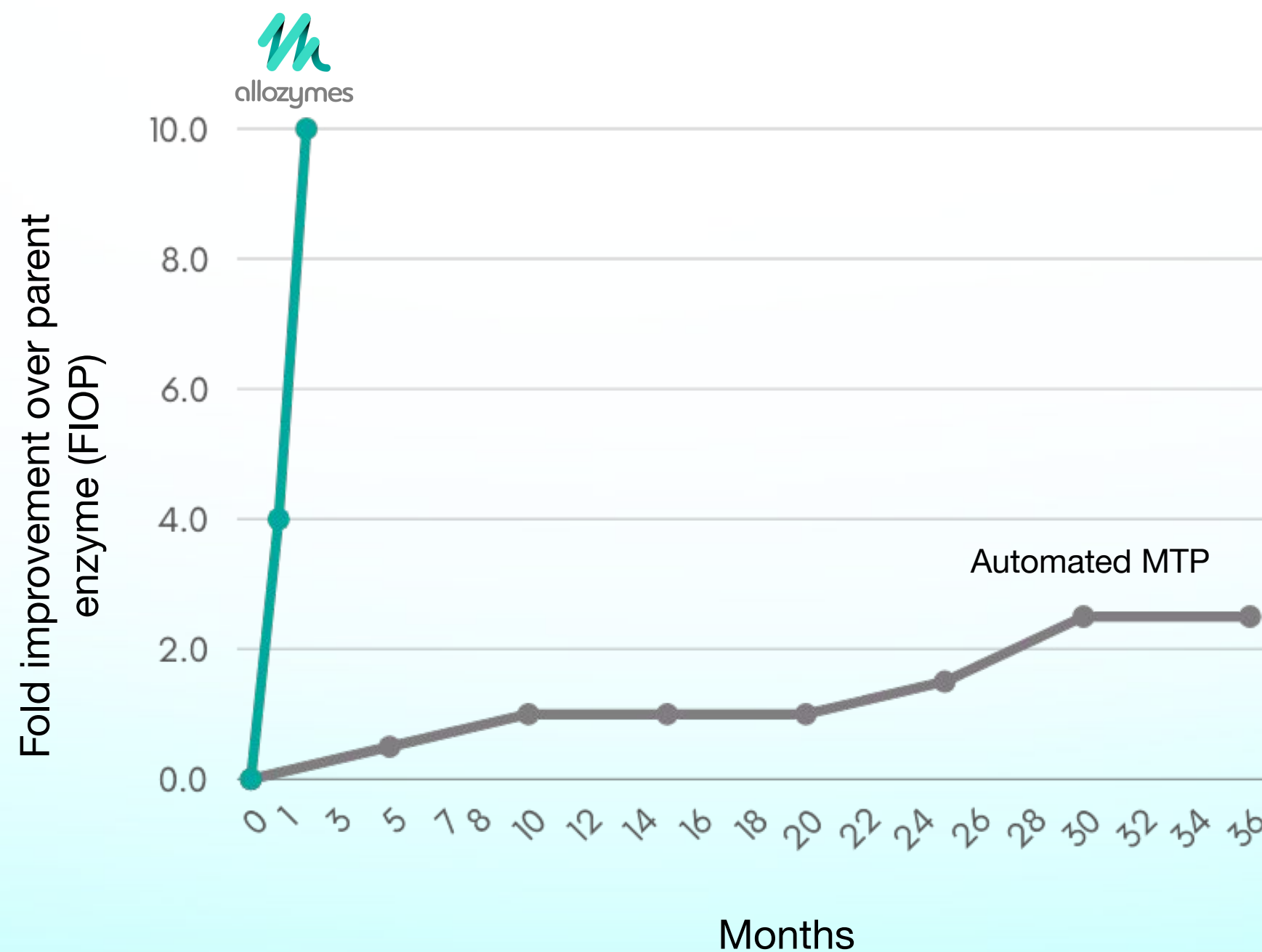


# Enzyme Activity – Improved Transferase Activity in 2 months

**Case outline:** Our customer had been working on the project for the past three years with limited success.



- Allozymes improved 10x activity in 2 months
- Generated >7,000 single variants and identified 5 hotspots
- Analyzed full combinatorial library of 3.2 million variants
- Delivered top variant with higher activity and ee >94%



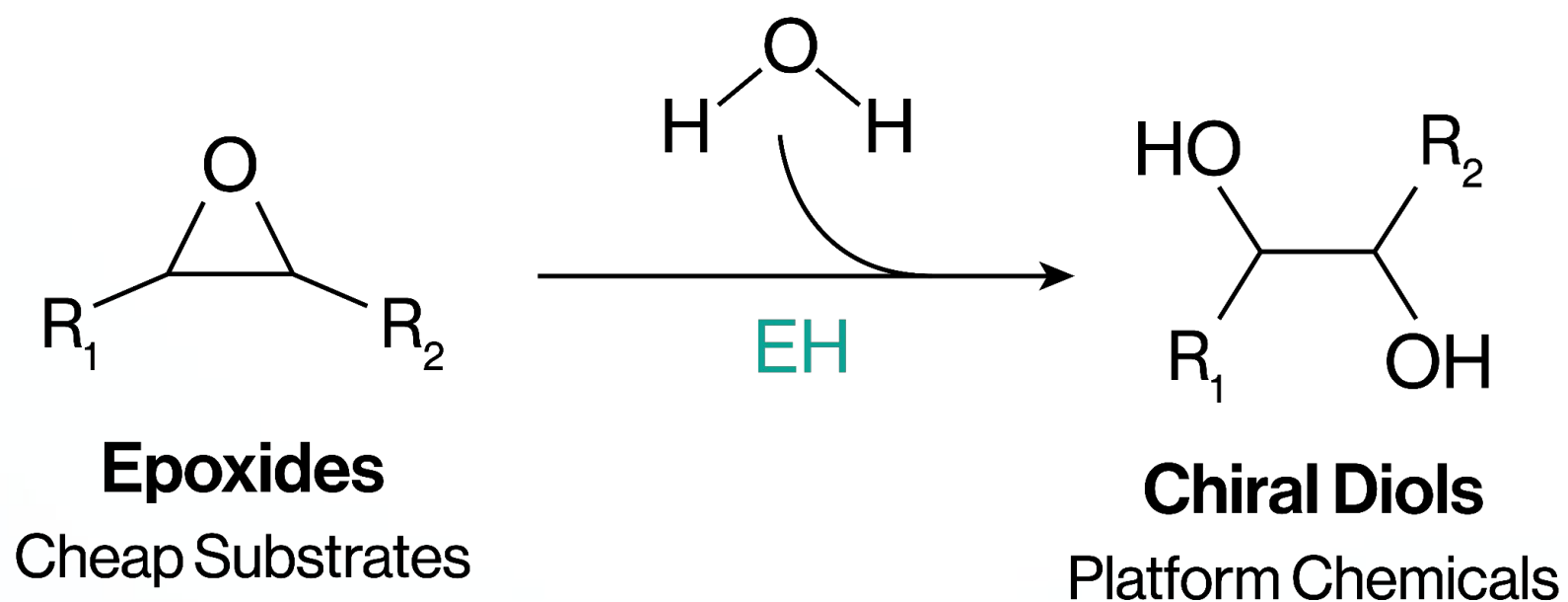




# Epoxide Hydrolase

**Case outline:** Producing diols from cheap epoxides

- Directed evolution by site-saturation mutagenesis on multiple positions was performed after analyzing crystal structure in 1 month



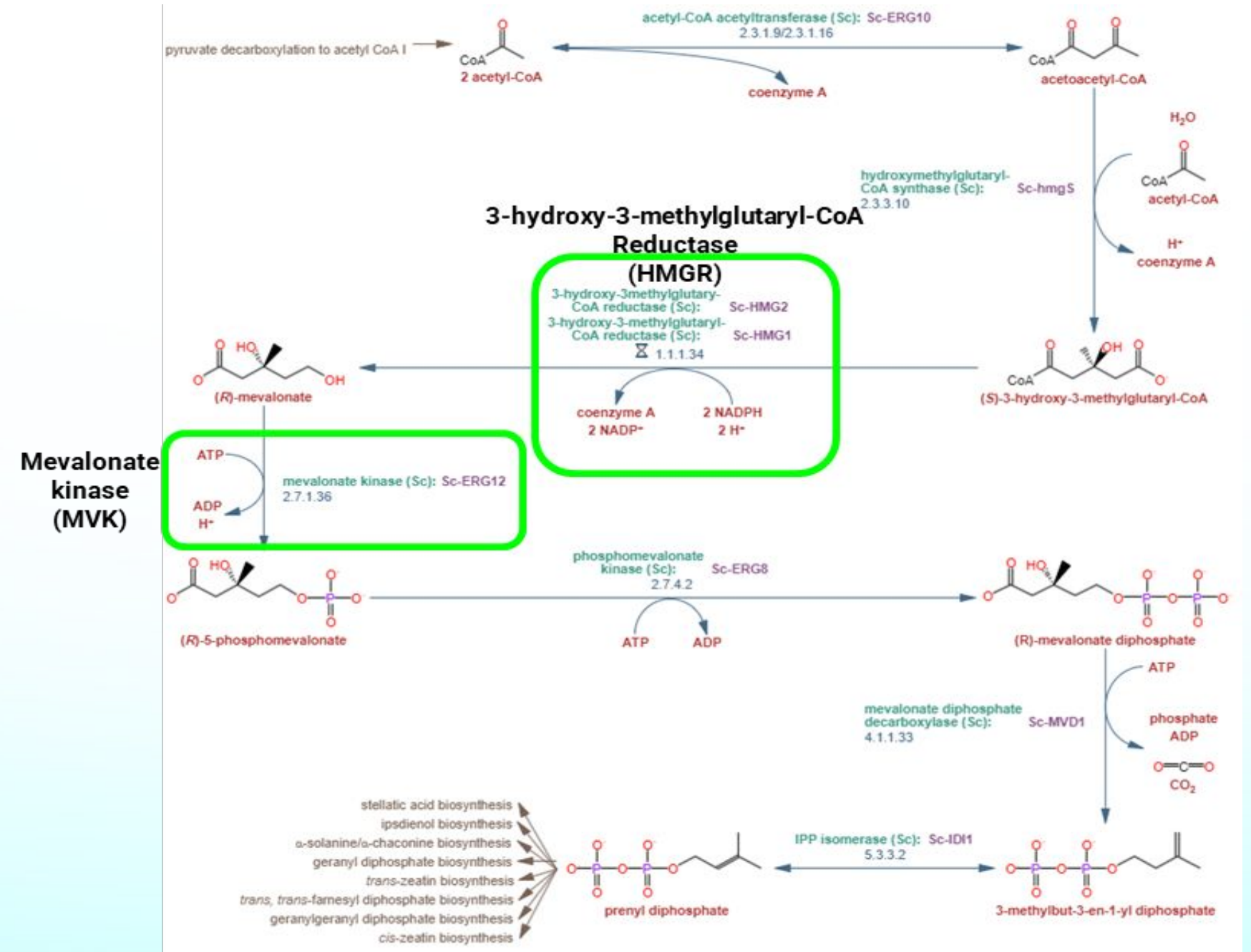
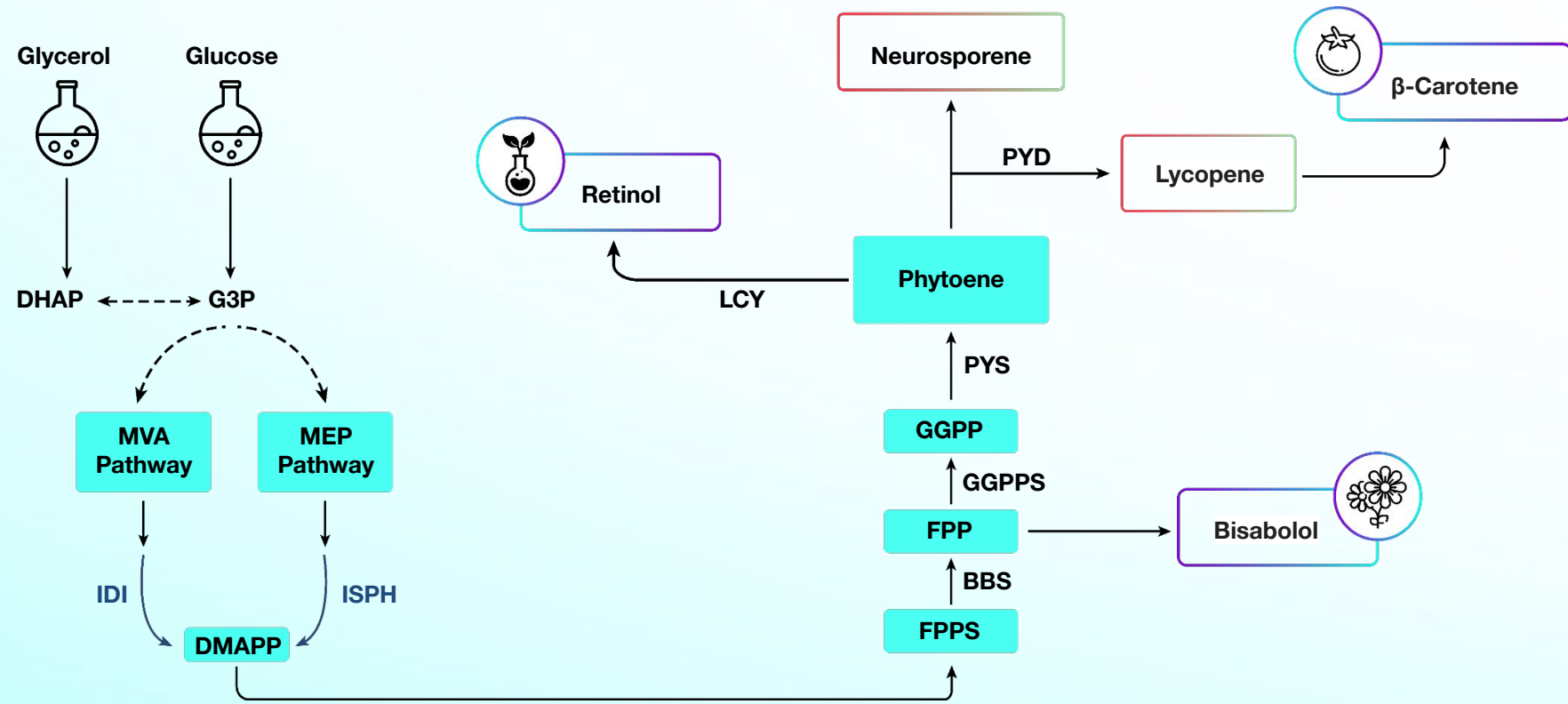
Screening round	Screened variants	Conversion (%)	Product ee (%)
1	32,800	65	86
2	98,400	90	95





# Metabolic Pathway

Case outline: Strain engineering for terpenoids and carotenoids from simple carbon source



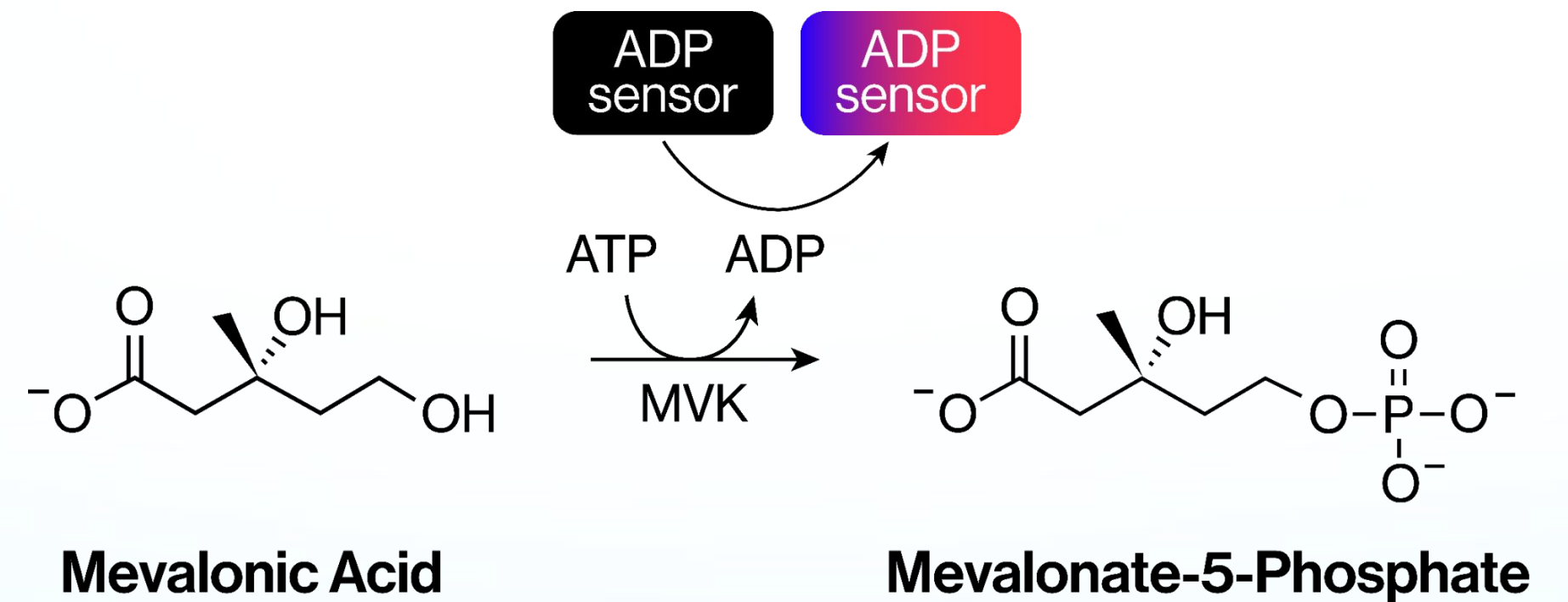




# Enzymatic Phosphorylation

**Case outline:** Engineer Mevalonate Kinase (MVK) to produce Mevalonate-5-Phosphate

- Kinase activity assay detects ADP produced during the conversion of mevalonic acid to mevalonate-5-phosphate
- High MVK activity → high ADP → More fluorescence
- Steps
  - Library creation
  - Intracellular enzyme expression
  - Droplet formation: Single-cell encapsulation with substrate
  - Pico-Injection of ADP sensor into the droplets
  - Detection and sorting of high fluorescence droplets







# Specialty Nutrition

## Increased decarboxylase activity in a metabolic pathway

**Case outline:** Our customer wanted to increase production of product which uses L-aspartate decarboxylase in pathway

- Cloning of target enzyme in *E. coli*
- Assay development (cascade reaction)
- Screening large enzyme libraries to identify hotspots
- Leveraged large datasets collected for data-driven learning to improve library design
- Performed iterative and enhanced screening campaigns to find best variants
- Evaluating up to 1000 variants within the strain

<b>Bottleneck enzyme</b>	Decarboxylase
<b>Enzyme Class</b>	Lyase
<b>Type of assay run</b>	Whole cell fluorometric assay
<b>Throughput &amp; Data generated</b>	Screened <b>42,000</b> variants to select <b>1000</b> variants, which were tested in upscaled assays.
<b>Outcome</b>	Improved activity <b>7x</b> in 7 months





Thank You