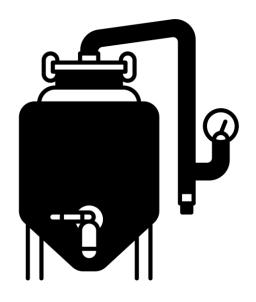








# Lecture #4 Fermentation Technology Overview



#### Today's agenda:

- Types of Fermentation
- Ingredients of Interest for Industry
- Current Investment Landscape
- Regulatory Environment
- Notable Collaborations

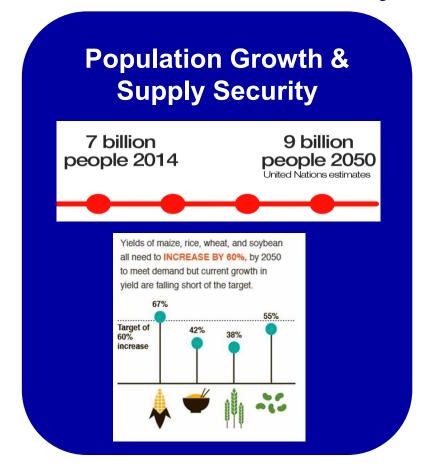


# Why Do we Need Fermentation Technologies?





"The world we want tomorrow starts with how we do business today" – Mars Inc. slogan





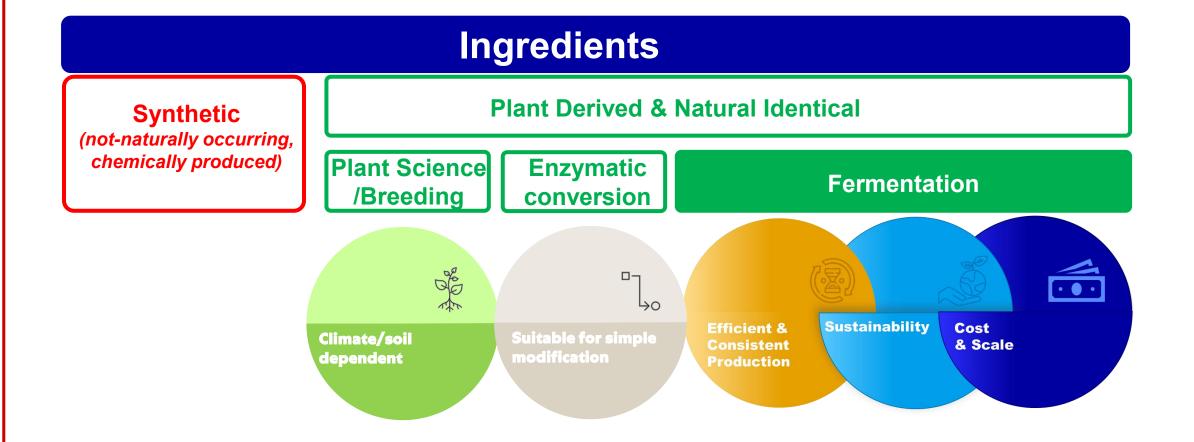






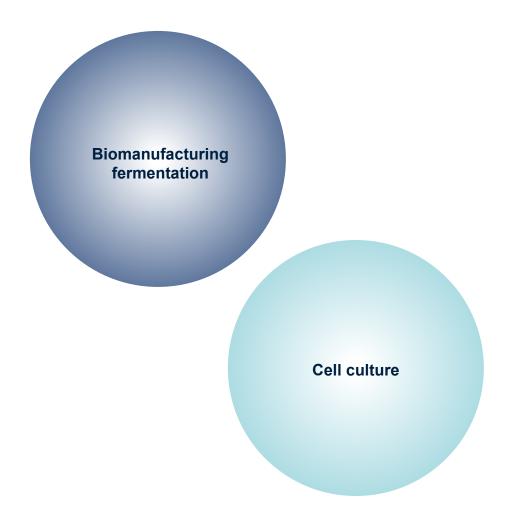


## Food Manufacturing in 2025 Must Utilize Traditional Agriculture and Technology

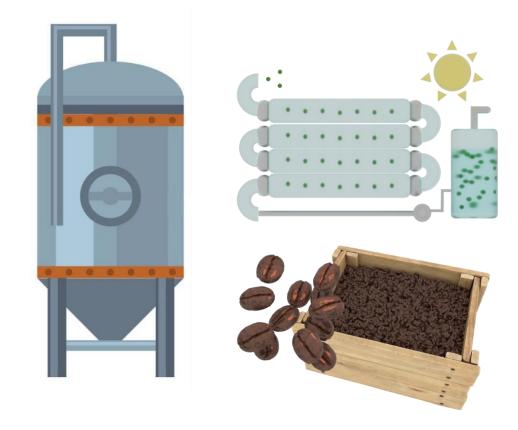


### For fermentation, context drives definition





• Fermentation in a **biomanufacturing context**: large-scale cultivation of microorganisms or single-celled creatures for industrial purposes

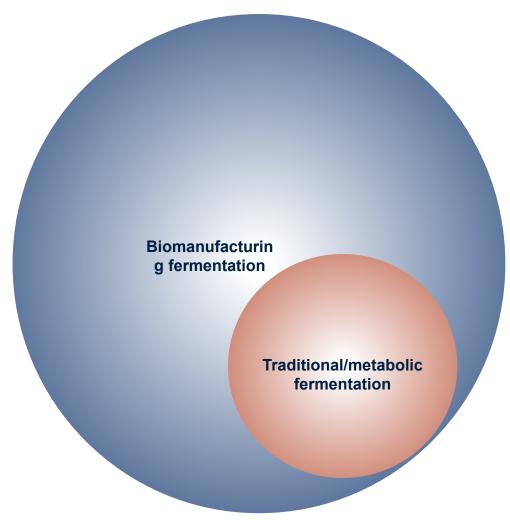


#### Traditional and metabolic fermentations overlap

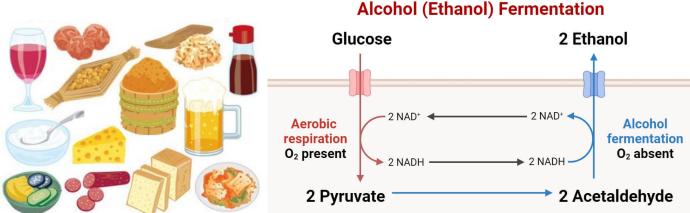






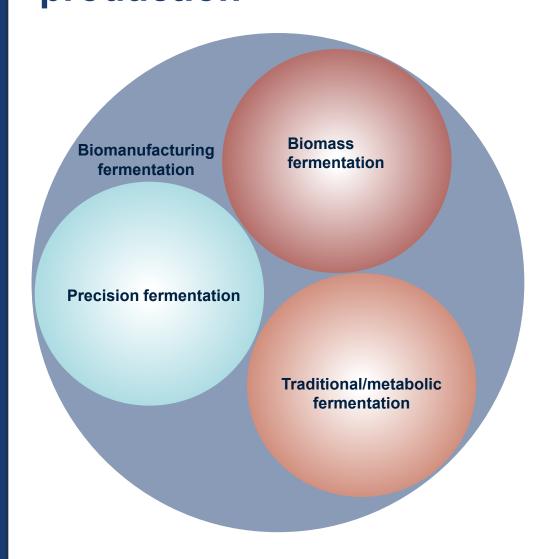


- Fermentation in a **biomanufacturing context**: large-scale cultivation of microorganisms or single-celled creatures for industrial purposes
- **Traditional** fermentation: yeast or bacteria generating ethanol, lactic/acetic acids, and CO<sub>2</sub> from fruit extracts, malted grains, and dairy
- Fermentation in a **metabolic** context: is an energy-generating process using organic compounds as both electron donors and electron acceptors

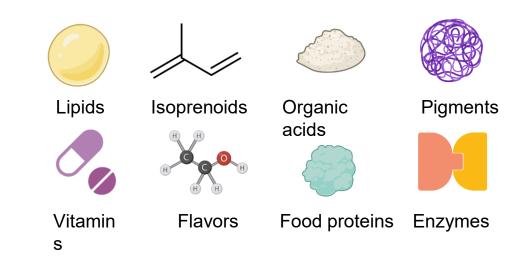


## Precision and biomass fermentation for food production





- Fermentation in a **biomanufacturing context**: large-scale cultivation of microorganisms or single-celled creatures for industrial purposes
- Biomass fermentation: the cells from GRAS organisms serve as the main ingredient of a food product or as one of several primary ingredients in a blend<sup>1</sup>
- Precision fermentation: uses microbial hosts as "cell factories" for producing specific functional food ingredients<sup>1</sup>



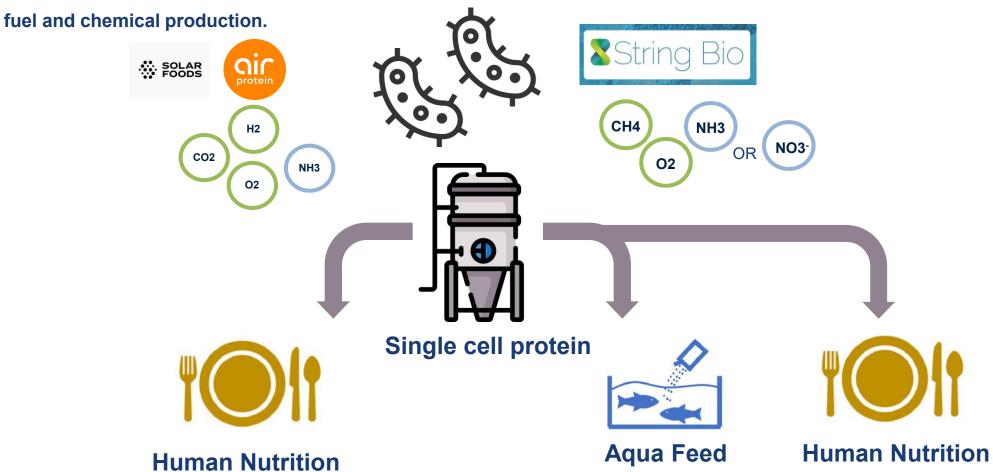
#### **Gas Fermentation**







Definition: A technology that utilizes carbon-fixing chemolithoautotrophic microorganisms to convert a variety of feedstocks, including industrial emissions and biomass-derived syngas, into a wide range of products, thereby supporting sustainable



#### BEZOS CENTER







Inputs

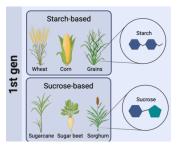
### **Typical Flow of Fermentation**



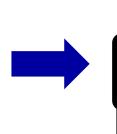
#### **Fermentation**

## **Downstream**Processing (DSP)

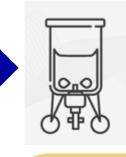
















Feedstock/Media
(Carbohydrate Source):
1st Gen: sucrose, glucose
Ag Byproduct feedstock

Microorganism: strain optimized for growth in certain media

- Time
- Temperature
- Shear

- Biomass Processing
- Centrifugation
- Filtration
- Pressing
- Purification

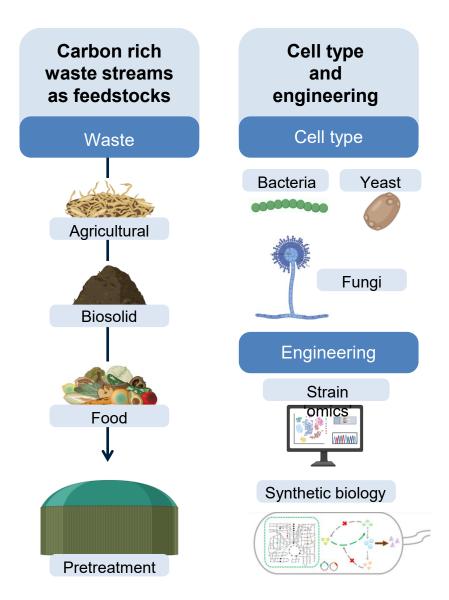
- Final Product
- Byproduct

## Generalized biomass/PF processes





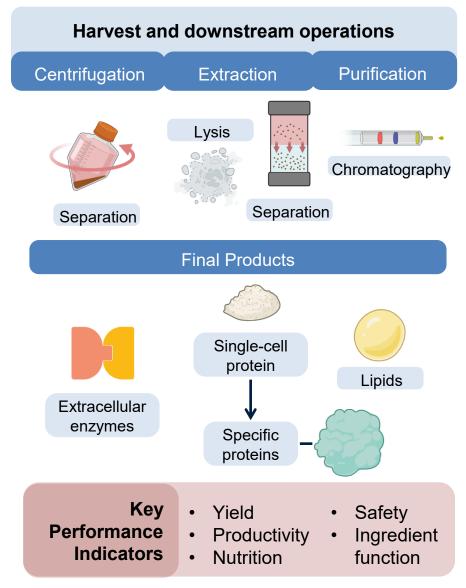




# Mode of operation Batch/fed batch Continuous

## Critical process parameters

- Temperature
- Dissolved Oxygen
- pH



## Bezos Center at NC State areas of targeted effort







Carbon rich waste streams as feedstocks

Screening

Cell type and engineering

Cell line dev

Manufacturing unit operations

Upstream

Downstream

Industry collaboration

Research collaborations

Bezos Center and BTEC at NC State University

**Product type** 

Macromolecules

Market factors, consumer acceptance technical and economic feasibility

Final product form

**Food Products** 

NC Food Innovation lab and Sensory Service Center

## Industry landscape of approved PF products







| Application                 | Company                    | Product   | Protein  | Strain                   | FDA GRAS Notice              |
|-----------------------------|----------------------------|---|--|--------------------------|------------------------------|
| Dairy                       | Perfect Day, CA, USA       | ProFerm™  | β Lactoglobulin  | T. reesei                | GRN No. 863                  |
|                             | ReMilk, Israel             | Animal -free milk and cheese  | β Lactoglobulin  | K. phaffii (P. pastoris) | GRN No. 1056                 |
|                             | Imagindairy, Israel        | Alternative milk and milk-based products  | β Lactoglobulin  | A. oryzae                | GRN No. 1145                 |
|                             | New Culture, CA, USA       | Animal-free cheese  | Casein   | L. lactis                |                              |
| Powdered proteins           | Helaina, NY, USA           | effera™   | <b>Bovine Lactoferrin</b>  | K. phaffii (P. pastoris) |                              |
|                             | Turtle Tree, Singapore     | Pink Gold™ (LF+)  | <b>Bovine Lactoferrin</b>  | K. phaffii (P. pastoris) | GRN No. 1219                 |
| Baking goods                | Onego Bio, Finland         | Bioalbumen <sup>®</sup>   | Ovalbumin  | T. reesei                |                              |
|                             | The EVERY Company, CA, USA | OvoPro <sup>TM,</sup> OvoBoost <sup>TM</sup>  | Ovalbumin  | K. phaffii (P. pastoris) | GRN No. 1104                 |
| Milk and dark<br>chocolates | OObli, CA, USA             | Milk and dark chocolates  | Brazzein, Monellin   | K. phaffii (P. pastoris) | GRN No. 1183 and<br>1142     |
| Meats                       | Impossible Foods, CA, USA  | Heme  | Soyleghemoglobin   | K. phaffii (P. pastoris) | GRN No. 1202, 737<br>and 540 |
|                             | Paleo, Belgium             | Myoglobin   | Myoglobin  | K. phaffii (P. pastoris) |                              |
| Cosmetics                   | Geltor, CA, USA            | Human Coll <sup>TM</sup> 2I,<br>Elastapure <sup>®</sup> ,<br>Caviance <sup>®</sup> , Collume <sup>®</sup> | Human type XXI collagen polypeptide, other collagen polypeptides | K. phaffii (P. pastoris) | GRN No. 1171                 |
|                             |                            | PrimaColl®  | Collagen   | E. coli                  |                              |

### Cost parity challenges in biomass and PF







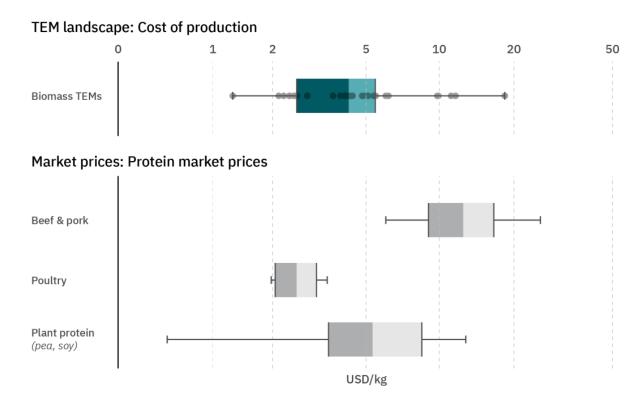


Figure 1. Biomass fermentation ingredient production cost (USD/kg); prices (in USD/kg) approach incumbent commodity ingredient market prices

According to GFI's latest TEM, biomass or single cell protein is closest to achieving cost parity to existing animal sources whereas there is a larger gap for precision fermentation.

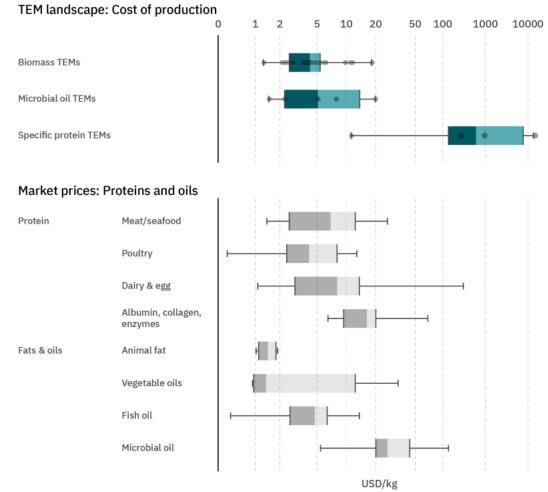


Figure 2. Biomass protein and microbial oil production costs highlight cost competitiveness with select incumbent ingredient market prices (USD)

## Improving value of microbial processes for food protein CR SUSTAINABLE PROTEIN

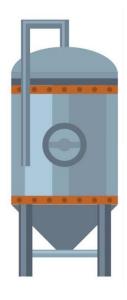
#### manufacturing





#### **Upstream bioprocess**

- Water
- Nutrients
- Seed inoculum
- Acid/Base
- Air (O<sub>2</sub>)



- Water
- Exhaust Gas (CO2)
- Cell mass
- Soluble product
- By-products (metabolites)
- Unconsumed medium components

$$A(C_6H_{12}O_6) + B(O_2) + D(NH_3) \rightarrow M(C_aH_bO_cN_d) + P(C_wH_xO_vN_z) + Q(CO_2) + R(H_2O)$$

Glucose

Carbon rich waste stream

E. coli cells

L. lactis SCP

Product

Yield = 2 g of glucose make 1 g of cells

Lactate

Acetate

Specific enzymes

#### **Notable Collaborations in Fermentation**







#### ADM joins with Air Protein to make meat analogs out of thin air

Published May 18, 2023 Megan Poinski







in 🖪 💥 🖶 🗷 🖫



**NEWS** 

26/3/2025

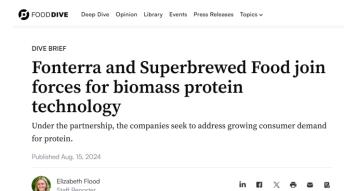
The Japanese food giant Ajinomoto continues to introduce new Solein®-powered products in Singapore

#### **AFN**

Fonterra/DSM backed startup Vivici unveils whey protein from fermentation: 'We are already at commercially viable titers'

February 19, 2024 | Elaine Watson

ALTERNATIVE PROTEIN



#### Cargill and ENOUGH expand partnership to provide consumers with innovative, sustainable protein options

MINNEAPOLIS (Feb. 15, 2024) - Cargill and food tech leader ENOUGH, which produces fermented protein sustainably, are expanding their current partnership to further innovate nutritious and sustainable alternative meat and dairy solutions consumers crave. Cargill is investing in ENOUGH's









## Thank you!