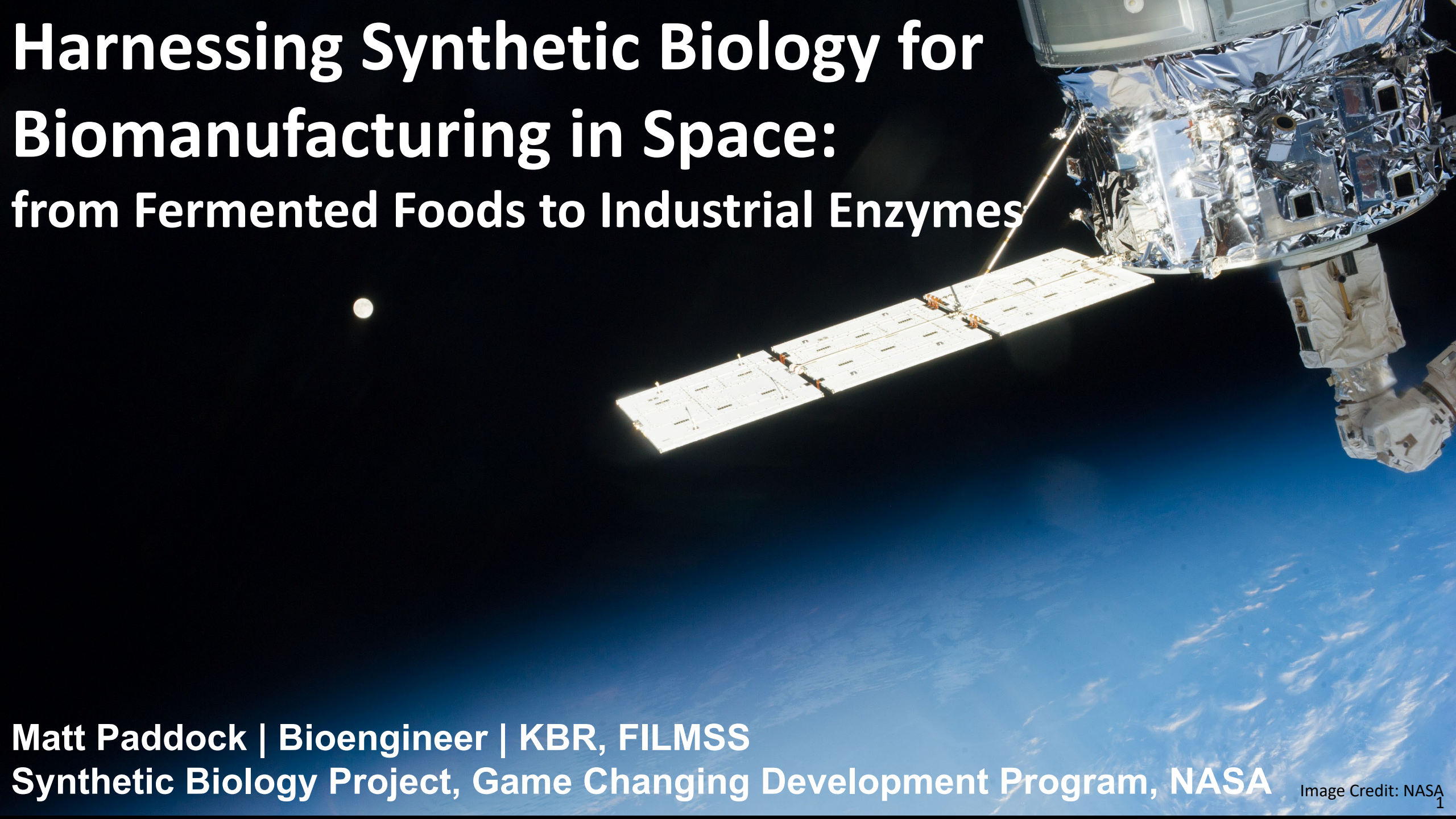
Harnessing Synthetic Biology for Biomanufacturing in Space:

from Fermented Foods to Industrial Enzymes



Matt Paddock | Bioengineer | KBR, FILMSS
Synthetic Biology Project, Game Changing Development Program, NASA

Image Credit: NASA

What is Synthetic Biology?

"Synthetic biology is a field of science that involves **redesigning organisms** for **useful purposes** by **engineering them** to have **new abilities**."

-National Human Genome Research Institute

Biological systems are:

- Scalable
- Programmable
- Precise (pure isomers)
- The only route of production in some cases (protein therapeutics)
- Low temperature and pressure
- Regenerable

Synthetic Biology in Space

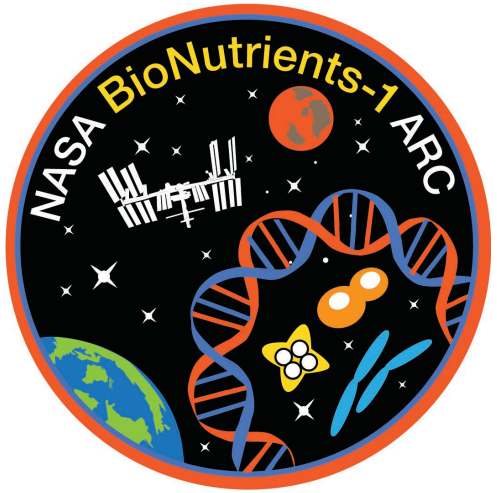
Mission resources will need to be produced on-site for mission sustainability/cost effectiveness

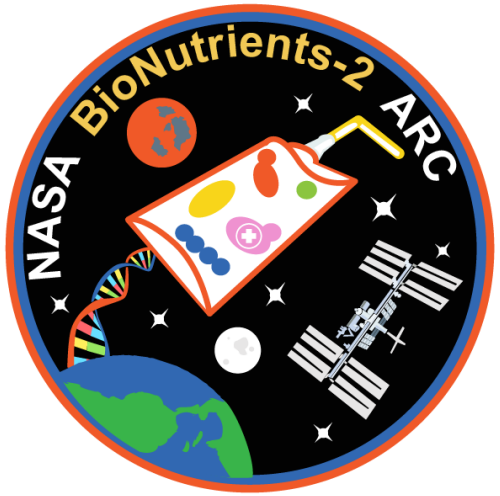
- Fuels, foods/nutrients, chemicals, plastics, binders, medicine, etc.
- Biomanufacturing can provide compounds abiotic systems cannot

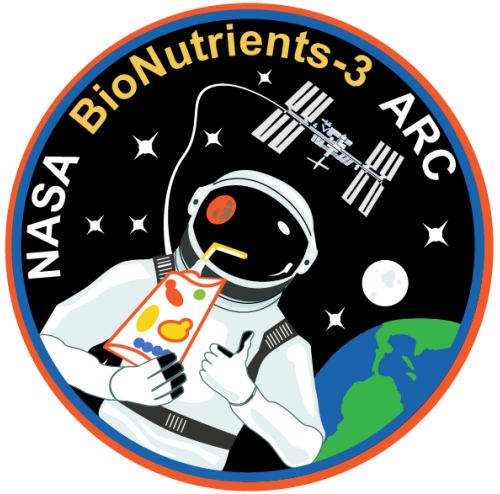
Image Credit: NASA

NASA Synthetic Biology Project

- Synthetic Biology Project
  - BioNutrients
    - Develop and demonstrate an on-demand nutrient production system for long duration missions to mitigate nutrient degradation in stored foods
    - Flight experiment on the ISS. Currently in third iteration







  - CO_2-Based Manufacturing
    - Develop and demonstrate a prototype system that enables microbial manufacturing via abiotic CO_2 conversion to products that drive biomanufacturing for future long-duration missions

BioNutrients Element: BN1, 2, and 3 Flight Missions

- Develop and demonstrate an on-demand nutrient production system for long duration missions to support nutrient degradation in stored foods.

Mission	Objectives	Launch
BN-1	5-year flight test Engineer yeast strains to produce carotenoids Test media and related packaging Develop Gen-0 bioreactor Test microorganisms for long-duration storage	2019
BN-2	<1-year flight test Expand products to yogurt and kefir Engineer medical countermeasure product Develop Gen-1 bioreactor to decrease system mass Develop HACCP food safety plan	2022
BN-3	Engineer system to produce multiple nutrients Develop Gen-2 bioreactor Demonstrate reliability of HACCP plan/Food Safety E-nose pathogen detection Pasteurization	2025

Image Credit: NASA

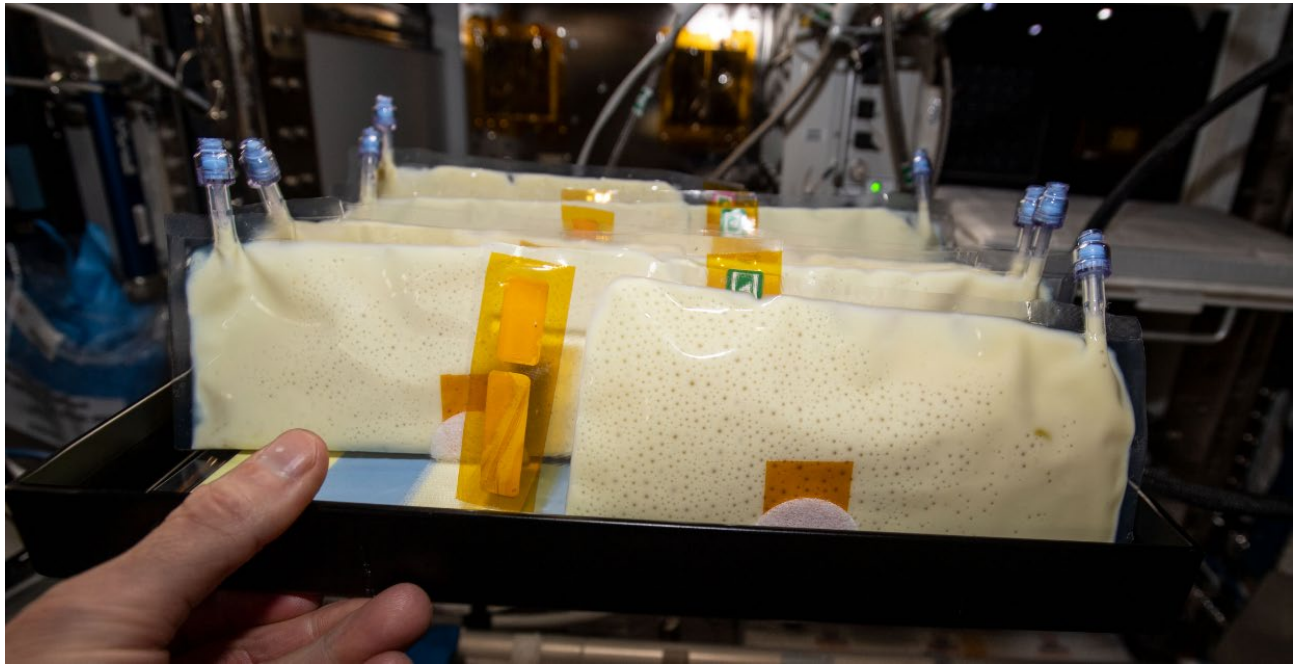
BN-2 Products

Microbe	GM Product	Purpose
Saccharomyces cerevisiae (Bakers Yeast)	Zeaxanthin	Supports eye health, may mitigate macular degeneration
Saccharomyces boulardii (Probiotic Yeast)	β-carotene	Vitamin A precursor
Kluyveromyces lactis (Dairy yeast)	Follistatin	Promote muscle formation to counteract microgravity effects
Streptococcus thermophilus (Yogurt bacteria)	Green Florescent Protein	Demonstrate genetic engineering of species
Streptococcus thermophilus *Lactobacillus bulgaricus* (YO-MIX 151 – Yogurt)	None	Test production and quality using a commercial organism, increase menu options
Lactic Acid Bacteria *Kluyveromyces marxianus* (C-FIR – Kefir Mix)	None	Test production and quality using a commercial organism, potentially increase menu options

24 h incubation pH indicator changes color to indicate yogurt is done.

Image Credit: NASA

BN-2 Operations on the ISS – no leaks, no anomalies, visible growth and yeast, yogurt and kefir production

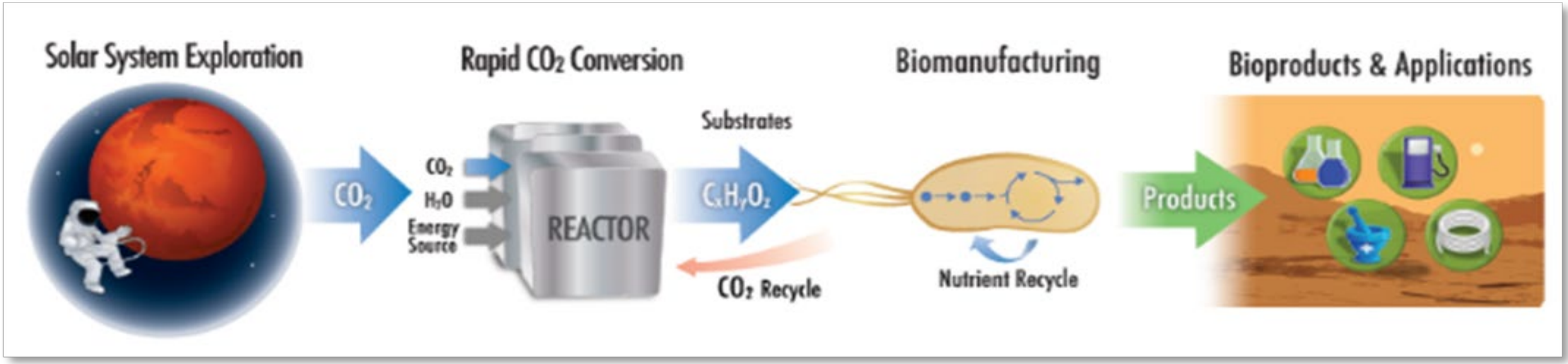


[Nicole Mann and Josh Cassada hydrating and shaking the yeast and yogurt packs, respectively. Featured in Space Station Science Highlights: Week of January 2, 2023 | NASA ISS Research on Twitter: "BioNutrients-2 tests an on-demand system to produce key nutrients from yogurt, kefir, and a yeast-based beverage. 🥛 The study could help maintain the crew's health while reducing launch mass and volume requirements. https://t.co/fuxW39uTWd https://t.co/bzFZVYMIxS" / Twitter]

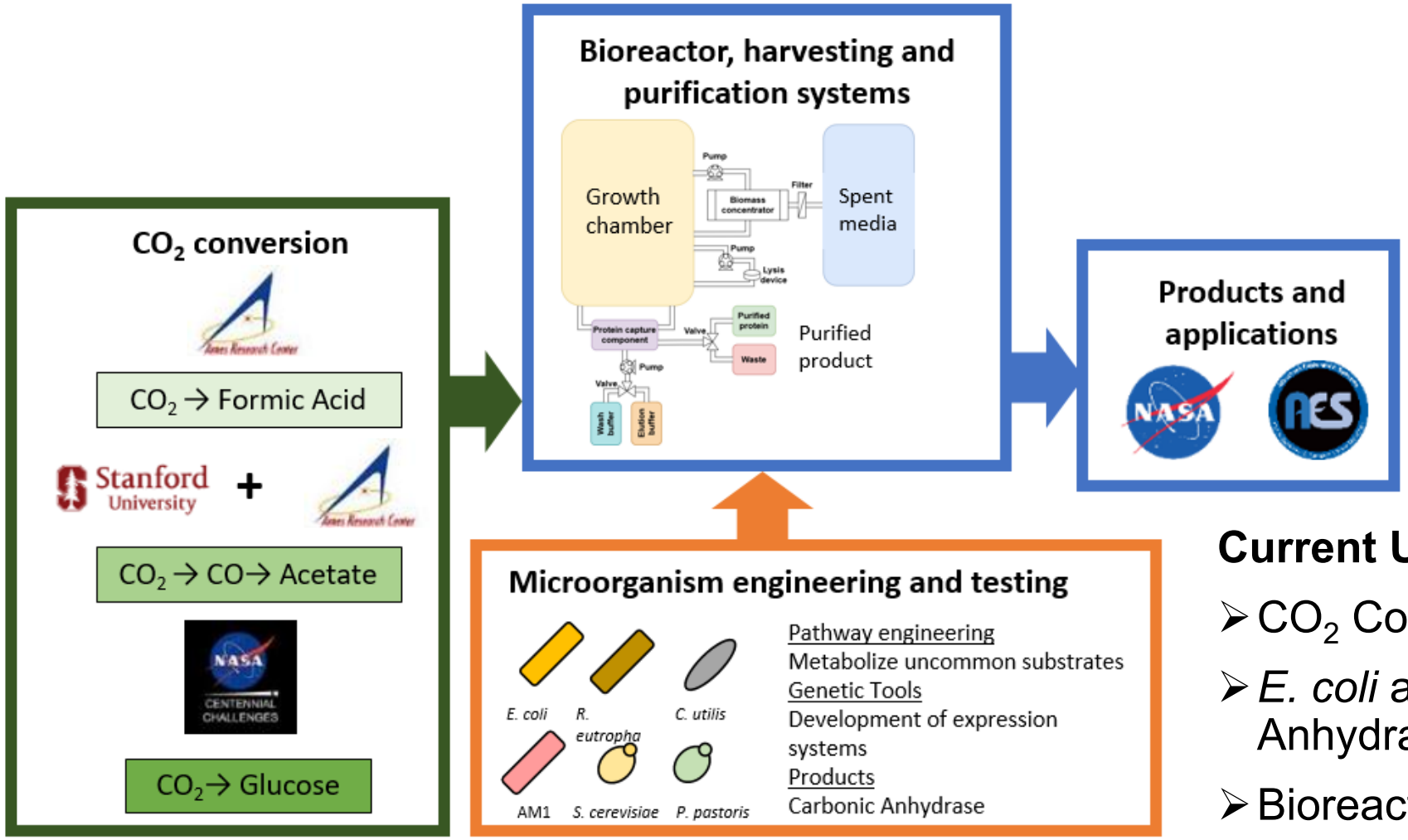
Image Credit: NASA

CO_2-Based Manufacturing

- Bioproducts are difficult to store and often need to be made on demand
- Commercial production systems are large, bulky, and not optimized for use on other planets
- Need to utilize local resources via In-Situ Resource Utilization (ISRU)



CO_2-based Manufacturing Project Element



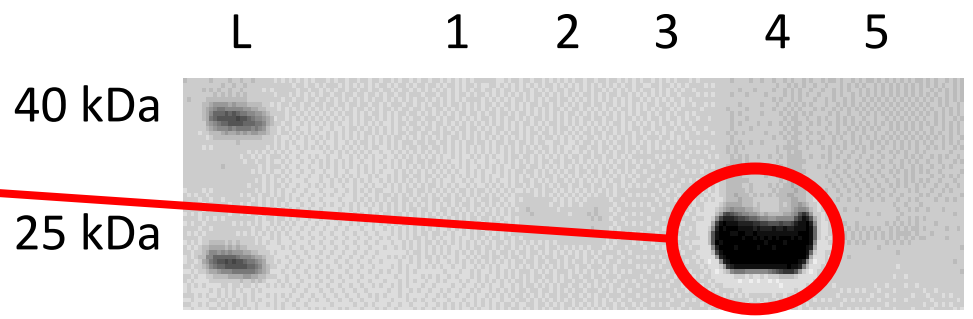
Current Use-Case Scenario:

- CO_2 Conversion to acetate and ethanol
- *E. coli* and *K. phaffii* to produce Carbonic Anhydrase and Cutinase
- Bioreactor and Harvesting Design
- Application for improved CO_2 removal in life support systems

CO_2-based Manufacturing Platform

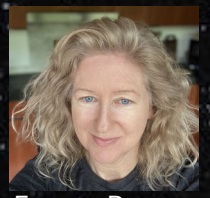
- Platform designed to be semi-autonomous to grow, harvest, dewater, lyse, and purify recombinant proteins from microbial cells
- Developed gravity-independent high-gas exchange membrane bioreactor to improve microbial growth
- Platform demonstrated validity of the concept and identified challenges for future investigation

High level of purification of recombinant protein



1. Supernatant, 2. Effluent from His-Tag Filter, 3. Wash Effluent, 4. Elution, 5. Post Wash Effluent

Synthetic Biology Team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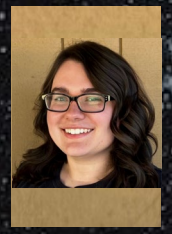
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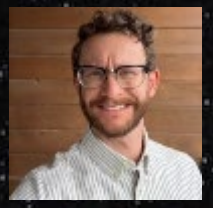
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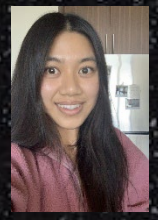
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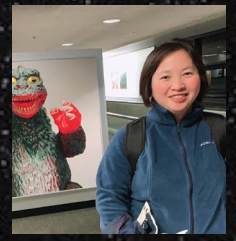
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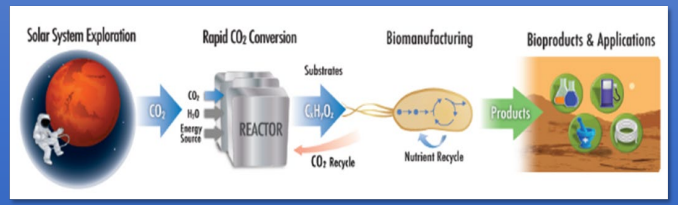
Q/A: Paul Milazzo
Logistics at KSC: Satro Narayan

Synthetic Biology GCD project Elements



CO_2 based manufacturing

BioNutrients

SPACE BIOSCIENCES
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Image Credit: NASA

Thank You

Questions?