



# Space Technology Mission Directorate Game Changing Development Program — Synthetic Biology (SynBio)

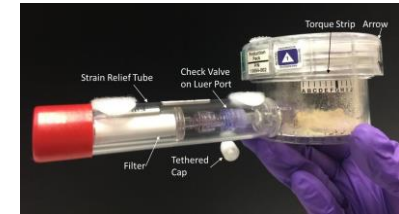
Frances Donovan | Synthetic Biology Project Monthly Status Report | 05.2021

# Project Overview



## ➤ Technology Product Capability

- Develop and demonstrate an on-demand nutrient production system for long-duration missions to mitigate demonstrated nutrient degradation in stored foods. Develop an evolvable platform for future surface missions – capable of producing other compounds (e.g., Medicines) requiring minimal resources. Perform an ISS time-course hydration, incubation, freezing and return for analysis over 5 year period.
- Develop and demonstrate a prototype system that enables microbial manufacturing via abiotic  $\text{CO}_2$  conversion to products that drive biomanufacturing for future long-duration missions.

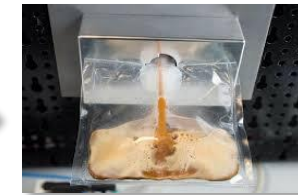


BioNutrients -1  
Flight  
Production  
Pack for on-  
orbit testing



## ➤ Technical Capabilities

- Reduced gravity compatible bioreactor development
- Long-duration ambient storage of microorganisms
- Methods for ensuring quality and safety of biomanufactured products
- Development of ISRU based growth media
- Space-relevant biomanufacturing system development
- Space qualified organisms for biomanufacturing

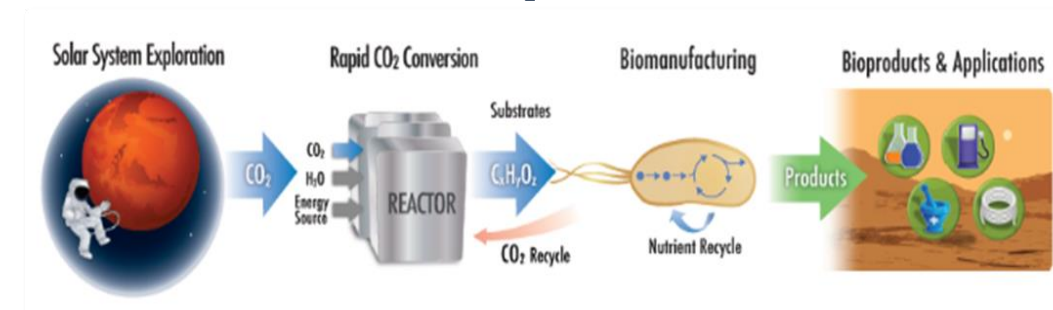


*Future Implementation Concept*

## ➤ Exploration & Science Applicability

- Cross-cutting: Supports crew health, enhanced logistics, mission sustainability and risk reduction for extended duration habitation missions on the moon (Artemis) and Mars and space-based outposts.

Hybrid  $\text{CO}_2$ -Based Biomanufacturing Concept





# Project Highlights/Accomplishments



Title	Type	Element	Description	New Image for EPO	Highlight Submitted
Systems Requirement Review	review	<b>BioNutrients</b>	Internal project milestone is complete. Science and Demonstration Requirements Document and Concept of Operations to be baselined week of 5/10/2021.	n/a	No
Phase 0/1 Safety Review	review	<b>BioNutrients</b> BN-2 mission	ISS Payload Safety Review Panel initial review of materials, chemicals, and operations proposed for BN-2 ISS mission. Panel had minimal concerns and provided feedback on expected safety verification tests and analysis. Status is approved with modifications of safety documents.	n/a	No

# SynBio



<b><i>Project description:</i></b>	Build organisms and bioreactor systems to demonstrate the value of emerging synthetic biology approaches to meet mission demands and support in situ resource utilization.		<b>Current TRL: 3</b>
<b><u>Cost:</u></b>  No cost issues at this time.	<div><div>G</div><div>G</div><div>G</div><div>G</div></div> <div>Monthly Trend</div> <div></div>	<b><u>Schedule:</u></b>  BioNutrients-2 production pack development and testing, and CO <sub>2</sub> -based manufacturing bioreactor and genetic engineering of organisms delayed due to mandatory telework. Lab access has been regained on a limited basis for BioNutrients activities only (Nov 30), and impacts to the schedule of both tasks are ongoing.	<div><div>Y</div><div>Y</div><div>Y</div><div>Y</div></div> <div>Monthly Trend</div> <div></div>
<b><u>Technical:</u></b>  No known technical challenges at this point, but schedule delays are impacting our progress.	<div><div>G</div><div>G</div><div>G</div><div>G</div></div> <div>Monthly Trend</div> <div></div>	<b><u>Programmatic:</u></b>  PPBE package submitted with new proposed milestone schedule due to COVID impacts. Minimal increase in cost (10k) in FY22, modest increase in out years. CR reflecting this request is in work.	<div><div>G</div><div>G</div><div>G</div><div>G</div></div> <div>Monthly Trend</div> <div></div>

# SynBio Additional Project Information



## BioNutrients

- BN-2 System Requirements Review (SRR) internal project milestone is complete. Science and Demonstration Requirements Document and Concept of Operations to be baselined week of 5/10/2021.
- BN-2 phase 0/1 Safety Review passed 4/28/2021.
- BN-2 Manifest Request Loader (MR Loader) data uploaded and ISS program review initiated on 4/29/2021.
- BN-2 Investigation Summary Form submitted for ISS program review on 4/30/2021.
- BN-2 Gen 1 bioreactor bag test with yeast engineered strains: Figure 1 biomass measurements indicate biocompatibility/growth and concomitant ethanol production within the bioreactor. This small amount of ethanol is expected and containment within the bioreactor is preferred by safety. Figure 2, Preliminary pressure testing of FEP bag indicates yeast growth will not create a pressure vessel. Preliminary testing indicates that the maximum design pressure of the bioreactor exceeds expected pressure values obtained during growth cycle. As planned, further testing and analysis will be used to verify safety requirements for this mission.

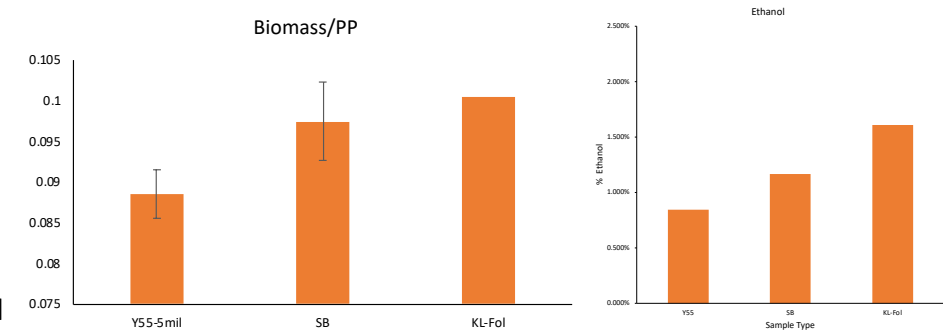


Fig 1 BN-2 Biomass results for Y55, Boulardii, K.lactis-FOL tested in 5mil FEP bags. Ethanol concentration in the bag following 48hrs of incubation. K.lactis wild type strain tested prior to testing with engineered follistatin producing strain.

## CO<sub>2</sub>-Based Manufacturing System Development

- Commercial support for test and fabrication as well as organism bio-engineering continues.
  - Purchase request in work to obtain a product-secreting capability in our chassis organism which will expand our capabilities and simplify product purification for secretion products.
  - Purchase request in work for a commercial vendor for proprietary cell lysis technologies to be tested for our system.
- Evaluation of commercial products and activities in the CO<sub>2</sub> based manufacturing space has led to ESM analysis of various COTS options.
- System requirements are being refined with systems engineering support, requirements document is being drafted.

## Project Management/General

- PPBE phase 1 package submitted. Proposes new schedule based on return to facilities July 1. CR is in work.
- TAPR and Design Review for BN-2 payload expected in June 14-25<sup>th</sup> time frame.
- CUBES Annual Review to be held May 5-6<sup>th</sup>.
- CO<sub>2</sub> conversion Centennial Challenge status: Phase 2 judging is commencing for three teams.
- Summer internships: two students have been selected for summer internships

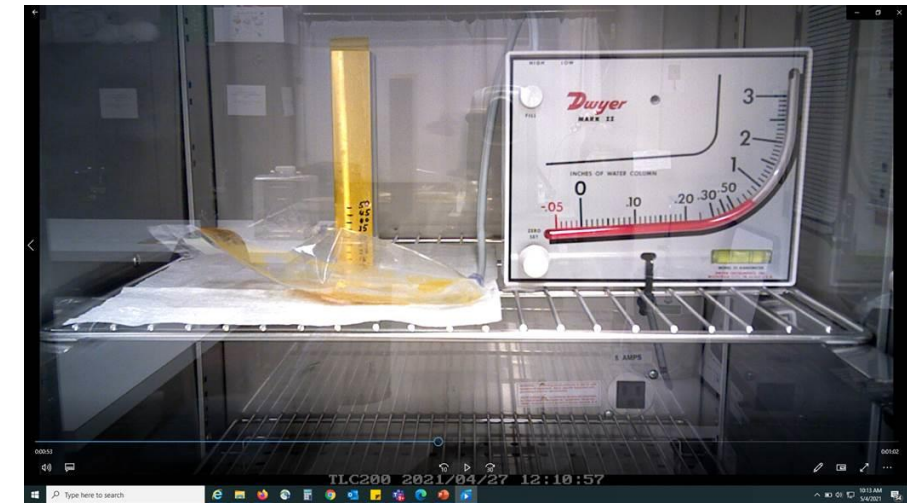


Fig 2 BN-2 FEP bioreactor growing yeast does not fully expand bag material and generates pressure far below the maximum design pressure. Preliminary results.

# Project Plan - Milestone Status



Milestone Title (Mirror Project Plan)	Baseline Date	Planned Date	Variance Explanation
<b><i>Status of milestones due since last report</i></b>			
C4-CO2 Space Compatible System Design Review	04/15/21	TBD, targeting Dec 2021	Delay due to lack of lab access, target date is for 6 months after expected return to lab. CR will be submitted in combination with PPBE per direction at PM forum.
<b><i>Status of milestones due in the next 60 days</i></b>			
C3-BN Generation 1 Flight Pack Design Review – will now combine with TAPR, targeting mid-June, new baseline set to July 15.	July 15	June 15-25 time frame	SME reviewers identified and their availability for this review confirmed. TAPR-ToR document is in preparation. Final date for review is dependent on panel availability but review will be virtual and we have reasonable confidence we can meet our target time frame.

# Project Risks, Issues/Concerns



Project Risks					
Risk ID	Affinity	Likelihood/ Consequence (if applicable)	Description/Status	Trend	Proposed Mitigations
Risk 1	S	5/2	If delays in lab access result in incomplete testing and data acquisition, the Design Review for BioNutrients-2 will not occur on Jan 19 <sup>th</sup> as planned.	realized	Delay the Design Review until the required testing can be completed. CR was approved to move review to be no later than July 15.

Issues/Concerns		
Issue Title	Description/Status	Proposed Mitigations
COVID-19 schedule impacts	The progression of several tasks on the project has been delayed due to lack of access to laboratory facilities. Upcoming controlled milestone, a design review for the BioNutrients-2 next generation bioreactor will not occur per plan and will likely have >30 day slip.	Delay the Design Review until the required testing can be completed. For other tasks the level of optimization and development will be impacted if schedule relief is unavailable.

# Education/Public Outreach



## EPO Involvement

- Paper submitted to 50th International Conference on Environmental Systems - ICES 202, July 12-14, team will attend virtually.

## EPO Calendar Outlook (High Priorities):

### 30 Day Look-Ahead

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### 60 Day Look-Ahead

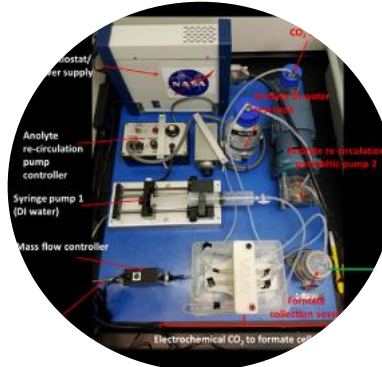
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# Space Synthetic Biology



View of BioNutrients-1 production packs during ISS crew operations. Packs were removed from incubator for mid-course agitation.



CO<sub>2</sub>-Based Manufacturing project prototype that demonstrated the production of formic acid from CO<sub>2</sub> and water, while also producing oxygen.



Ground testing of BioNutrients-1 production packs in SABL flight incubator (ground unit) to verify temperature, fit and no leakage.



View of open BioNutrients-1 production pack during loading. Dried edible media (powder) and yeast pellets (red) are visible.



State-of-the-Art CO<sub>2</sub> electrochemical system that converts CO<sub>2</sub> and water to formic acid without electrolytes. The formic acid serves as a media for microbial production systems.



Loading BioNutrients-1 production packs with media and organisms under sterile conditions. The pack lid and sterile water filter are attached during this assembly.