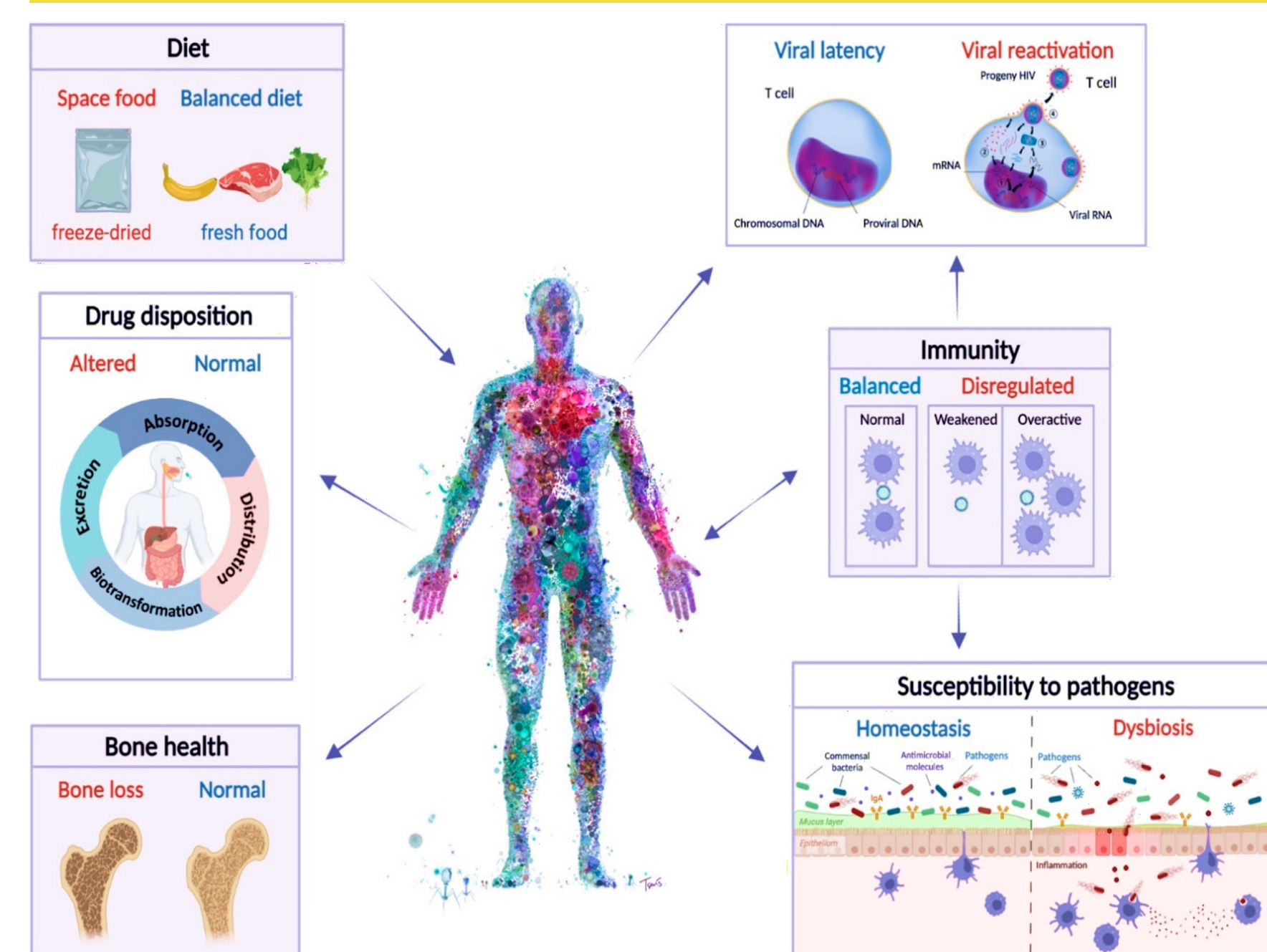


## Introduction



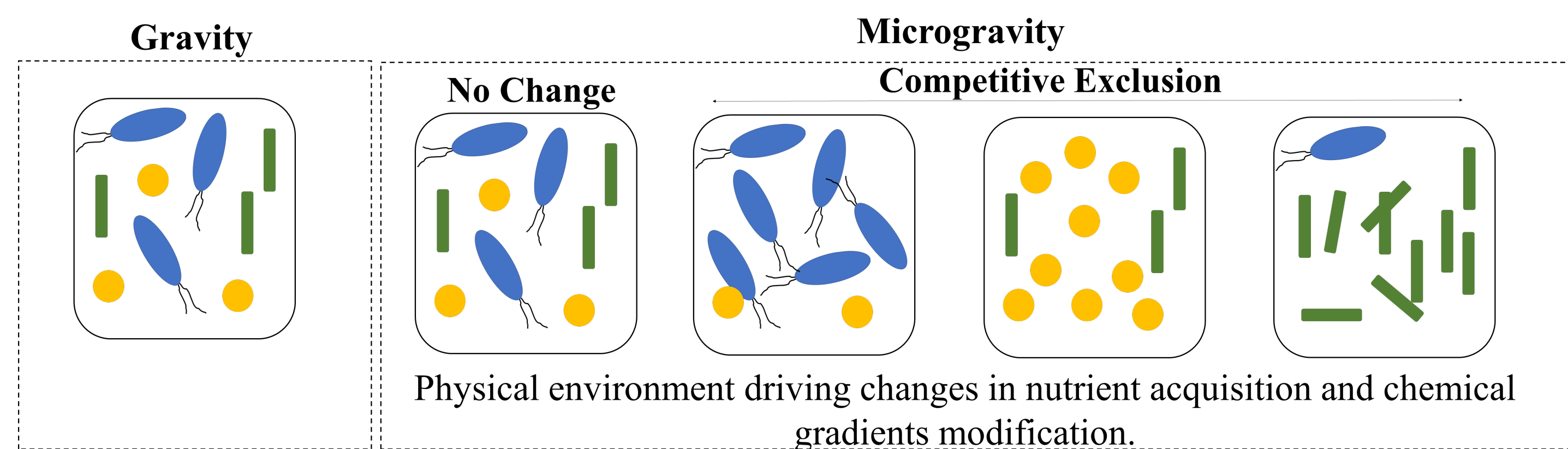
- NASA is establishing a lunar base to prepare for increased human space exploration
- Extended human space operations
- Increased exposure

\*Image Credit Tessei, D., et al., Life, (2022)

to space stressors such as microgravity ( $\mu\text{G}$ ) can result in significant cellular and physiological changes

- Space travel has transient and long-term impacts on astronaut microbiome that alters immune system function
- Research studies that evaluate microbial ecology under  $\mu\text{G}$  are limited

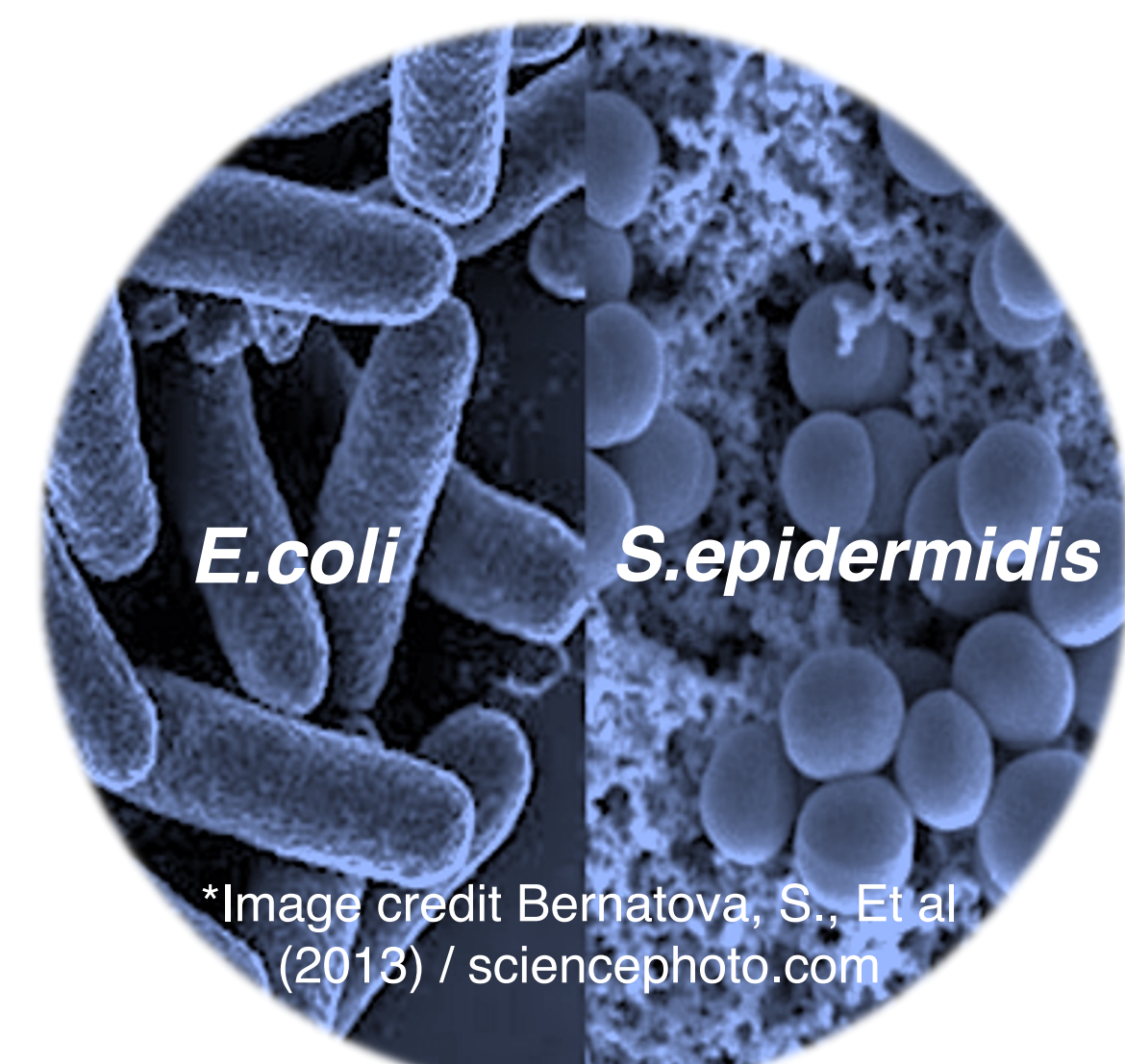
## Hypothesis



- $H_1$ : Microbial communities develop atypical phenotypical characteristics and growth patterns under simulated  $\mu\text{G}$  environment compared to nominal gravity
- $H_0$ : Microbial communities demonstrate typical gravity phenotypical characteristics and nominal growth curve

characteristics under sim  $\mu\text{G}$  environment

- Viable Colony Formation Units (CFUs) comparison
- Phenotypical evaluation
- Differential gene expression

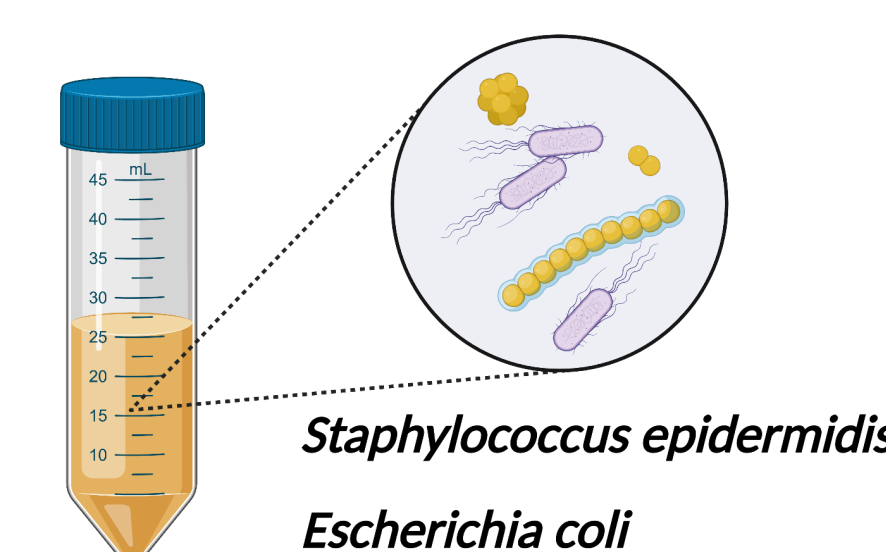


\*Image credit Bernatova, S., et al., (2013) / sciencephoto.com

## Experimental Design

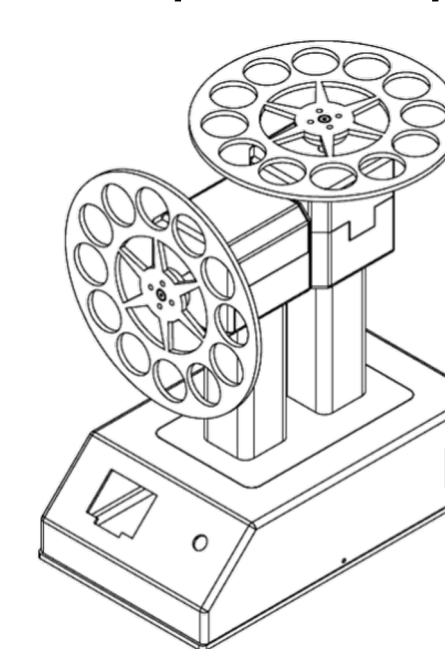
- Escherichia coli* (Intestinal Microflora) – Gram Negative & Rod Shaped
- Staphylococcus epidermidis* (Epithelial Microflora) - Gram Positive & Cocci Shaped
- Eosin Methylene Blue (EMB) media – Gram Negative
- Mannitol Salt Media – Gram Positive

1 1:1 Mixed Colony Inoculum

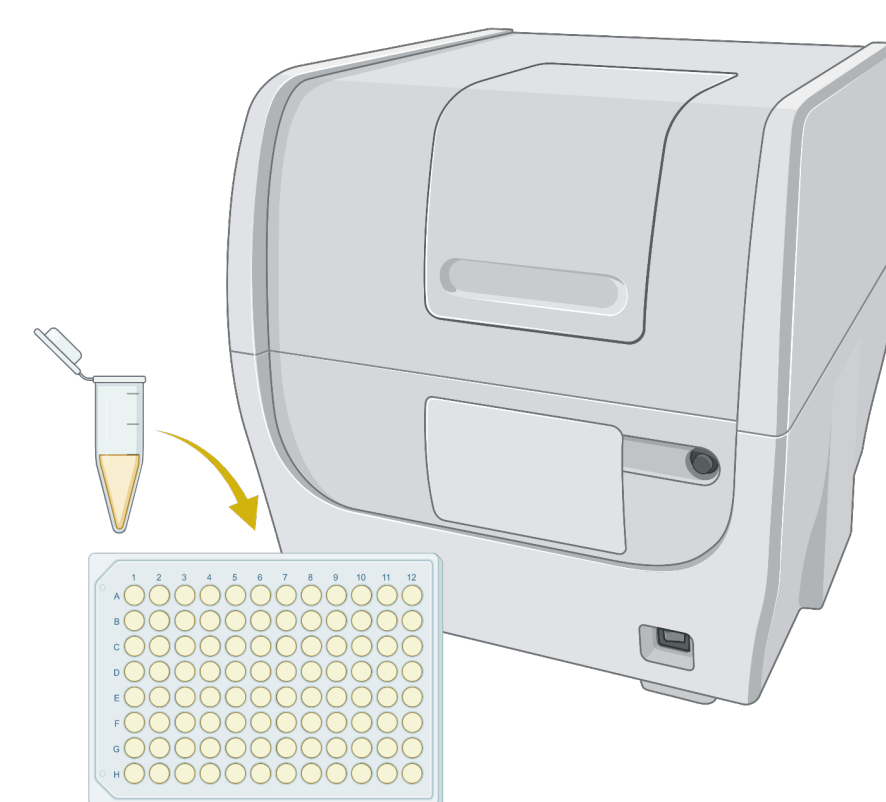


2 Simulated  $\mu\text{G}$  Exposure via 2D Clinostat (EagleStat)\*\*

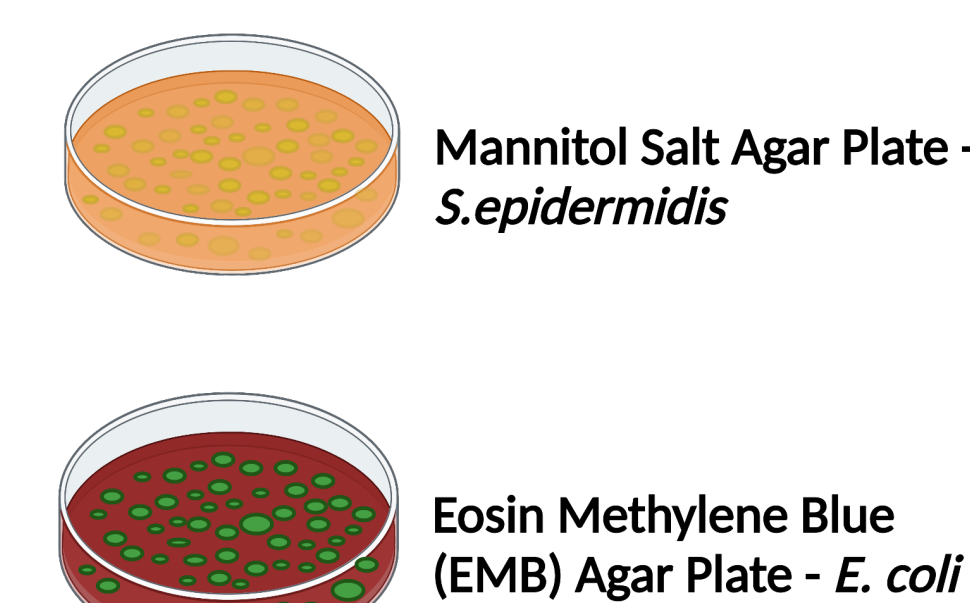
\*Samples pulled hourly for 21 Hours.  
\*\*24 Hour sample run independently



3 Cell Density Measurement via Spectrophotometry ( $\text{OD}_{630\text{nm}}$ )



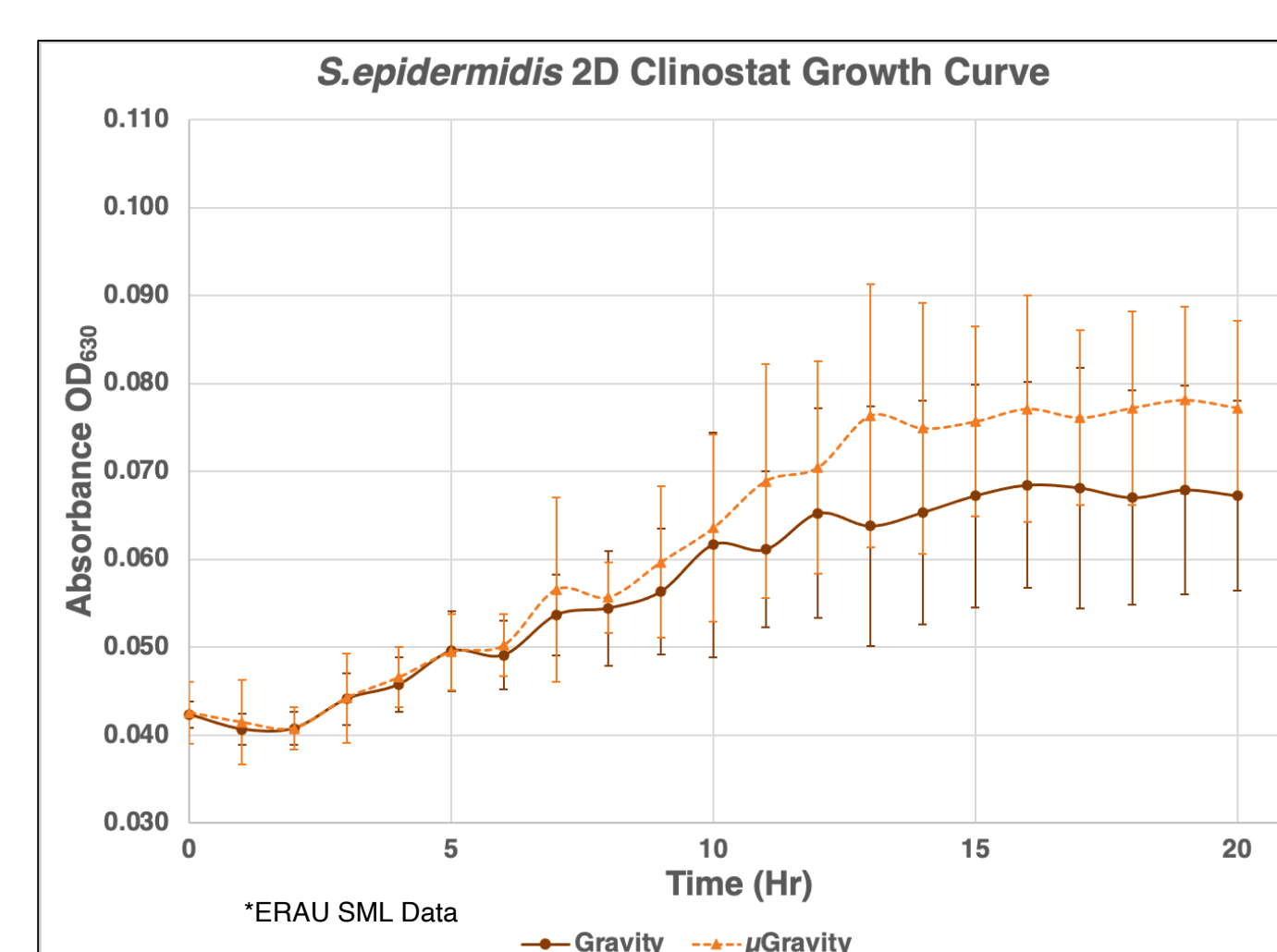
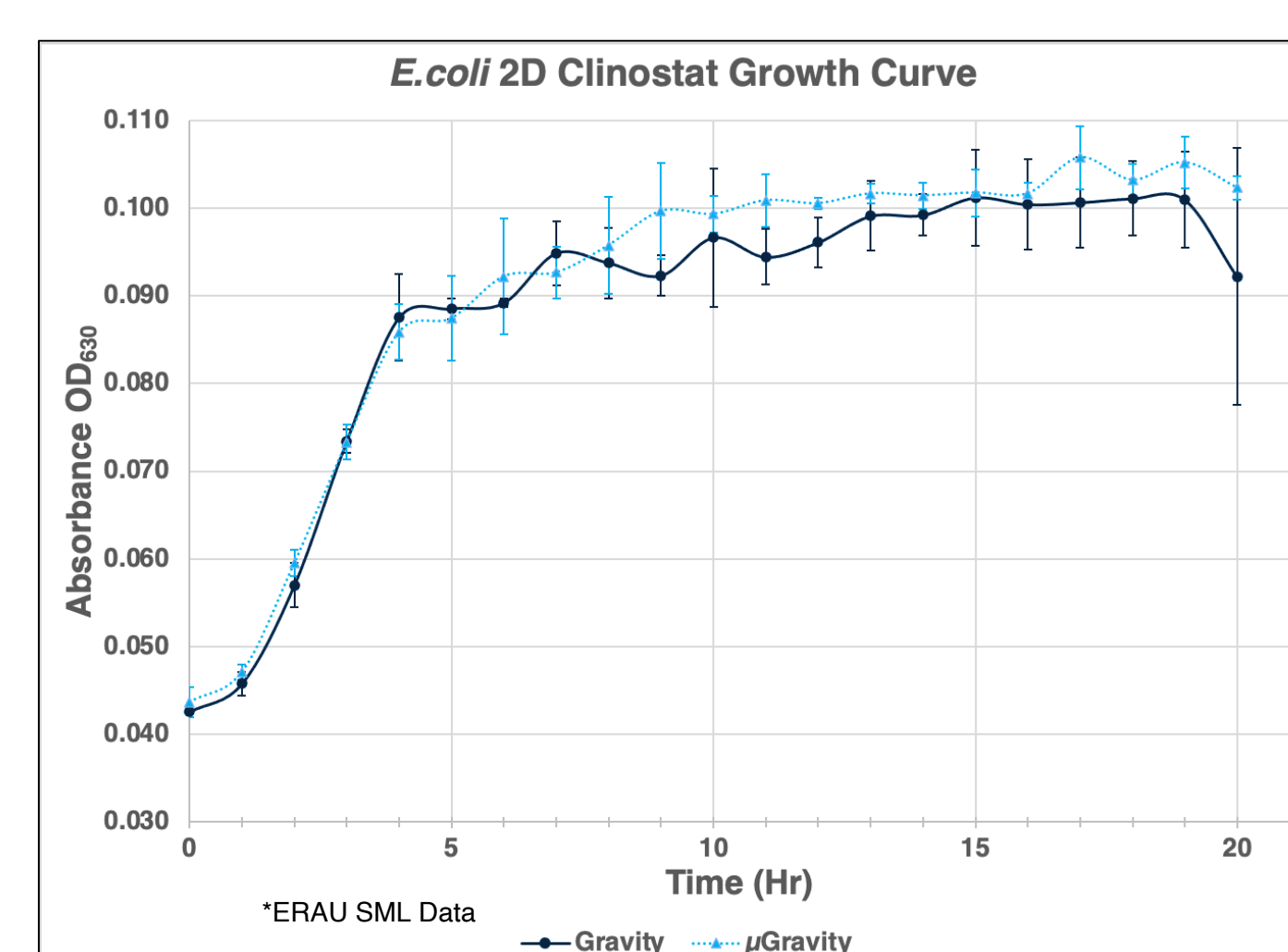
4 Serial Dilution and Drop Plating to Determine CFUs



\*Image Generated using BioRender

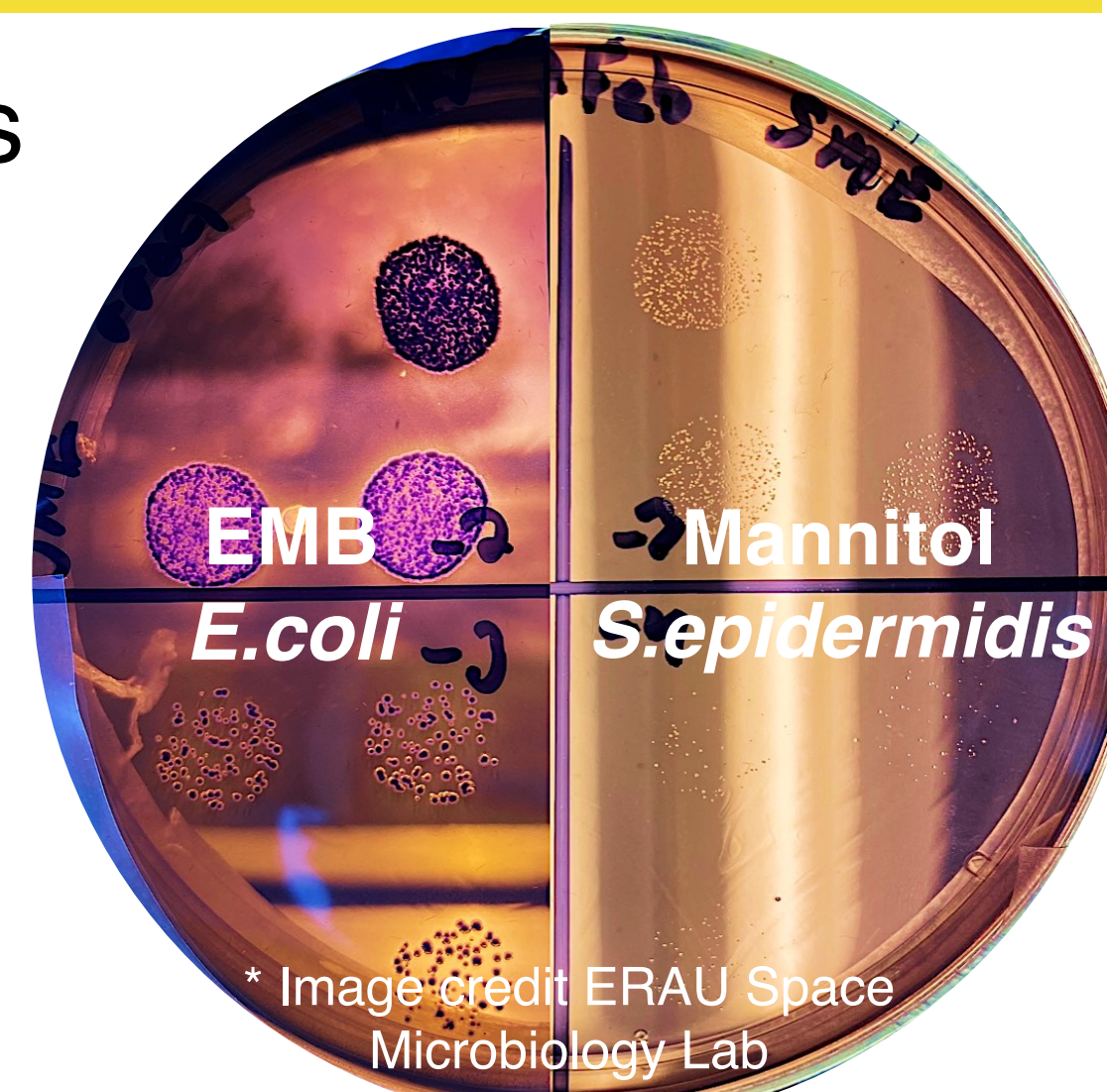
## Growth Curve Results

- 6 Clinostat Trials = 252 Samples (3 Biological Replicates)
- Spectrophotometry measurements completed in triplicate per sample at optical density of 630nm
- Slight increase of cells under simulated  $\mu\text{G}$  compared to gravity analog

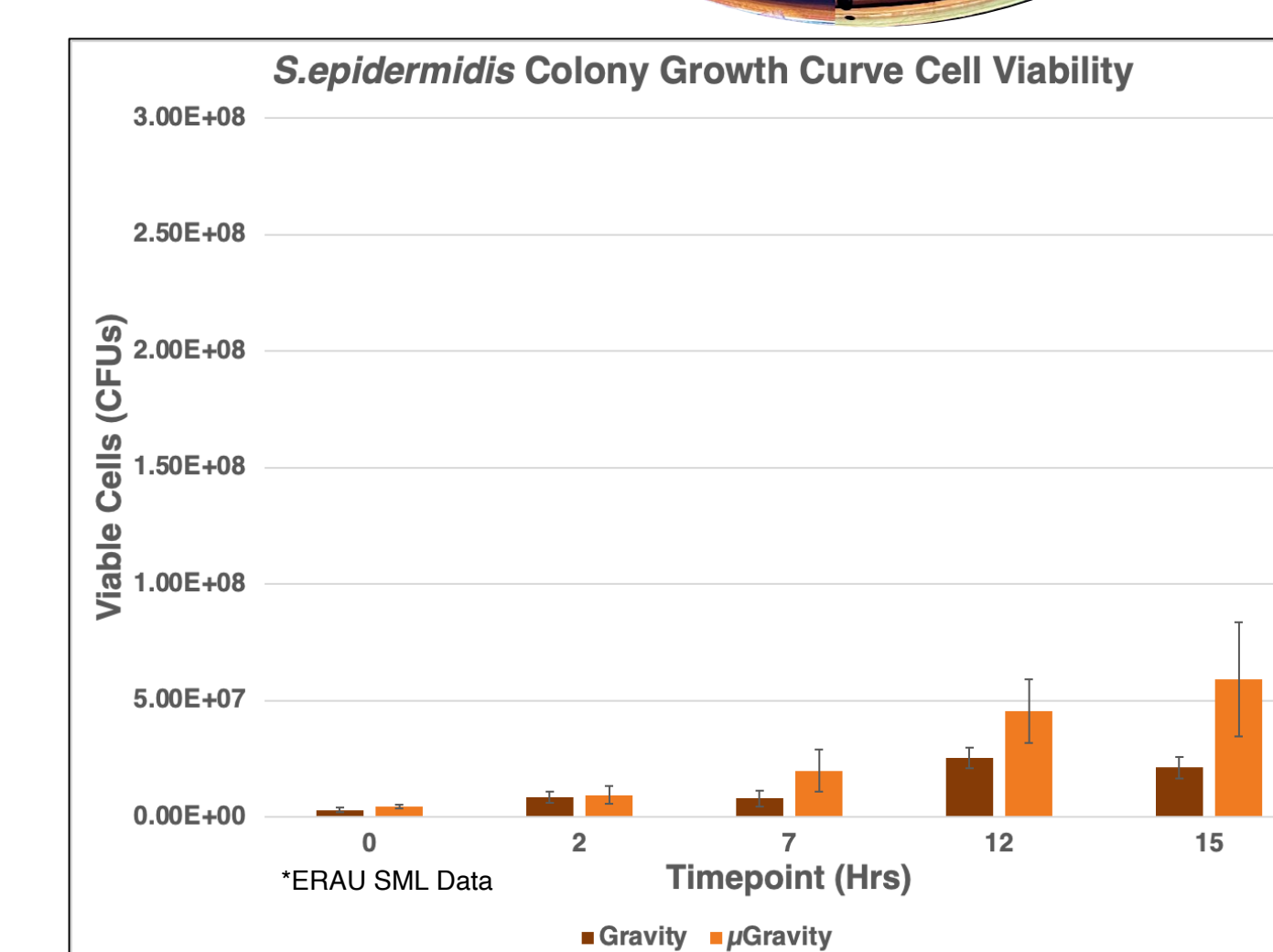
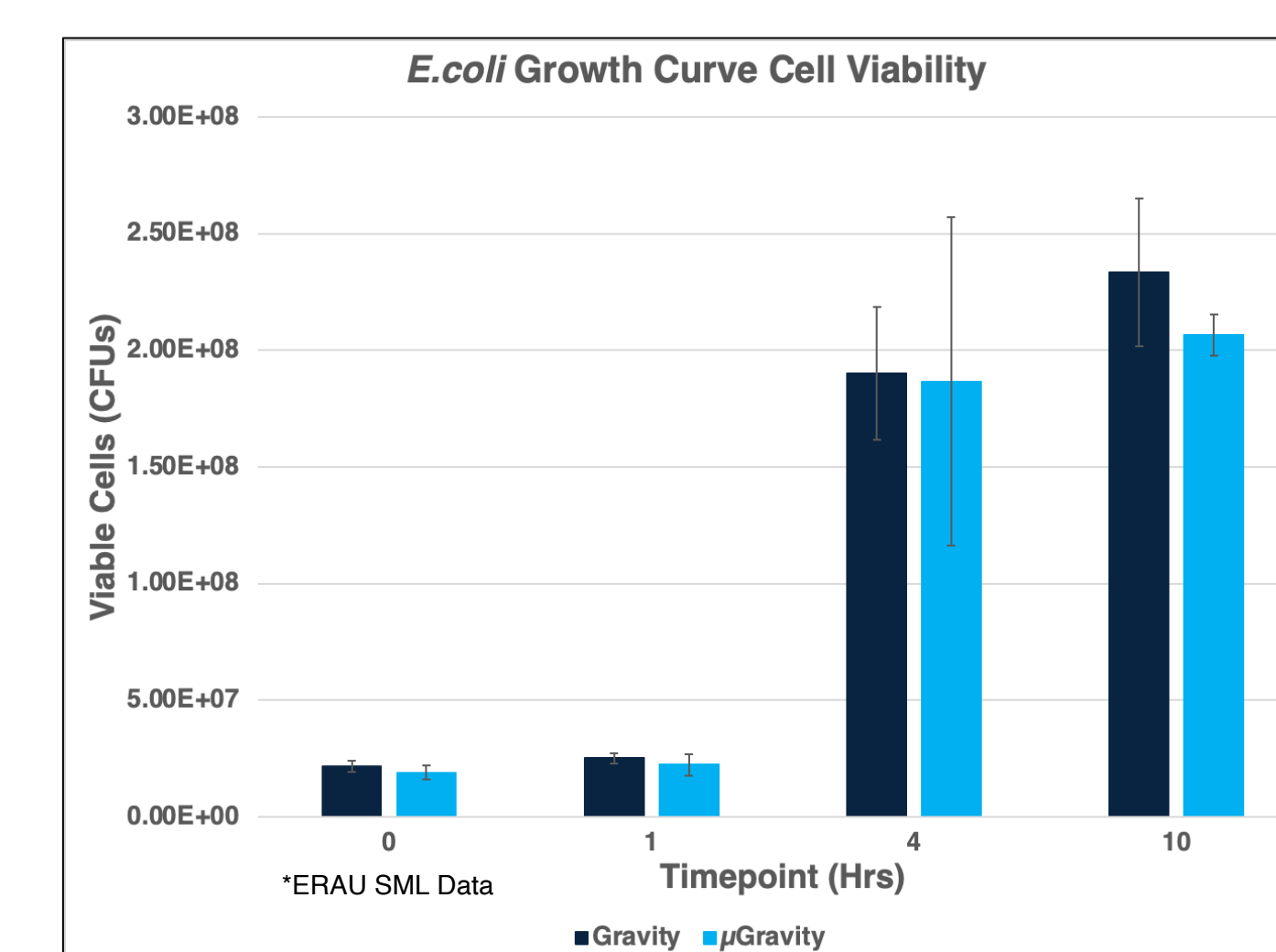


## Growth Curve Viable Cells Results

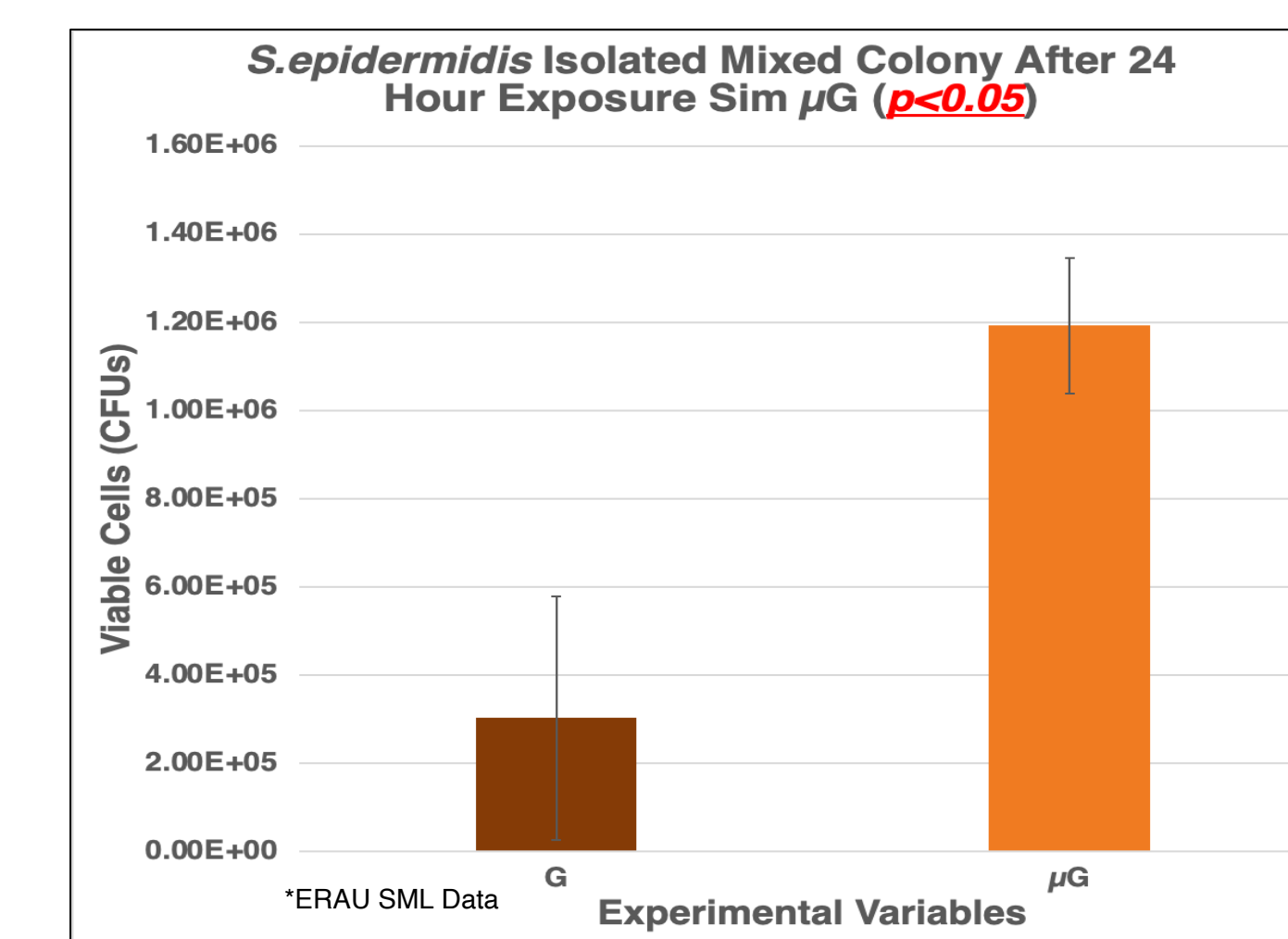
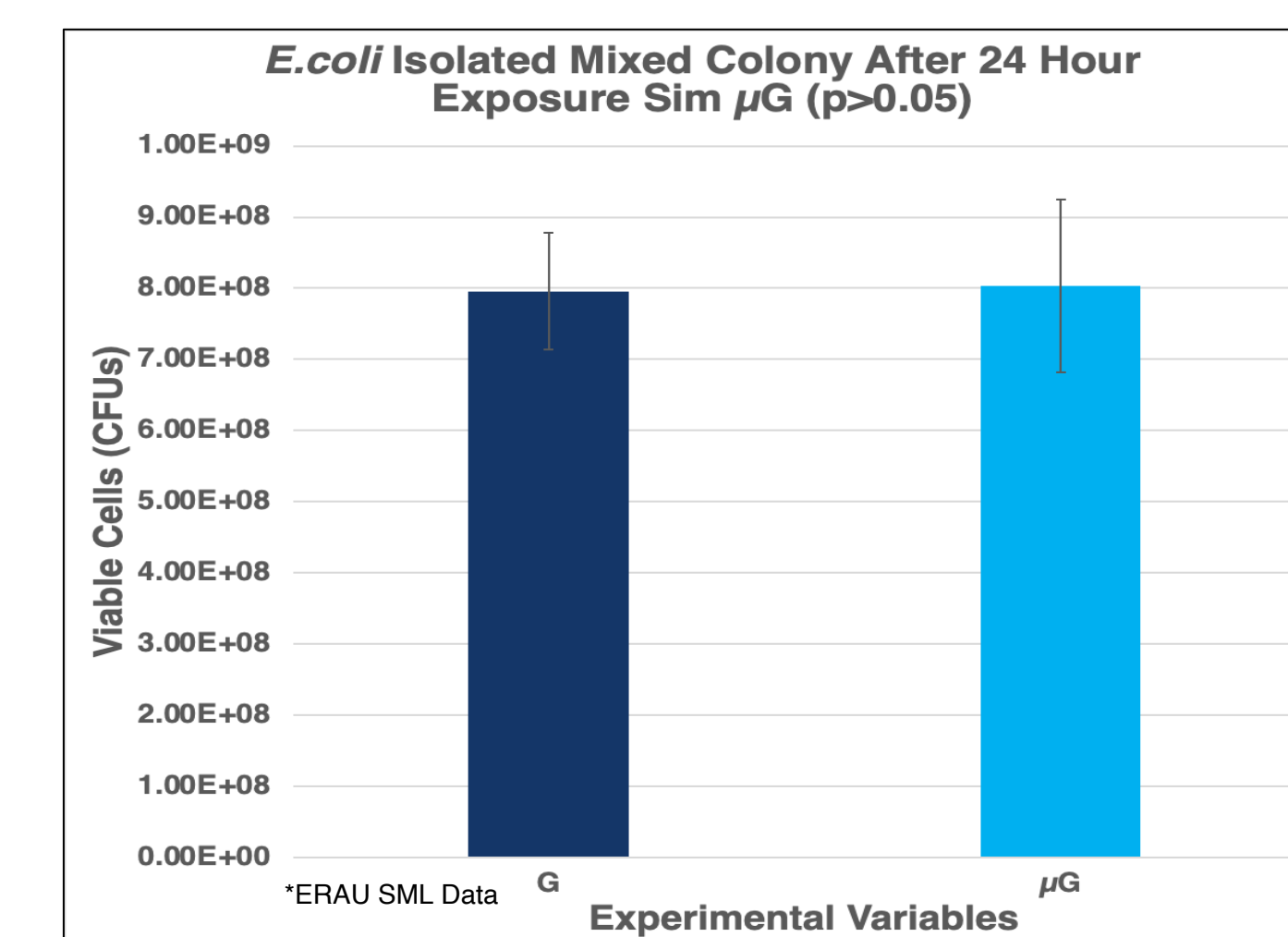
- 11 Timepoint Samples – 22 Plates (3 Biological Replicates per)
- Timepoint samples selected from growth curve phases
- S. epidermidis* Hour 15  $p > 0.05$



\*Image Credit ERAU Space Microbiology Lab



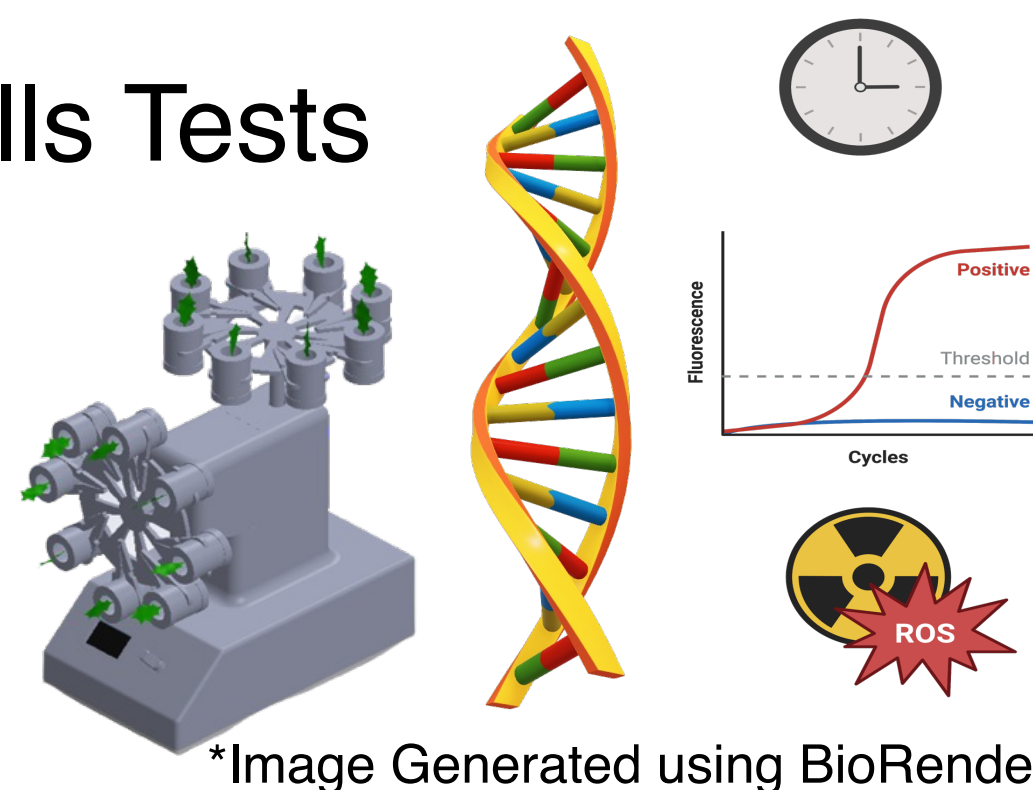
## Mixed Culture Results



- Significant ( $p < 0.05$ ) viable cell increase (~1.5 fold increase) with *S. epidermidis* under simulated  $\mu\text{G}$  conditions

## Future Research

- Timepoint Mixed Culture Viable Cells Tests
- Differential Gene Expression
- Low Dose Ionizing Radiation
- Lunar Regolith Soil Communities



\*Image Generated using BioRender

## References

Kuehnast, T., Abbott, C., Pausan, M. R., Pearce, D. A., Moissi-Eichinger, C., & Mahner, A. (2022). The crewed journey to Mars and its implications for the human microbiome. *Microbiome*, 10(1), 1–14. <https://doi.org/10.1186/s40168-021-01222-7>

Tesei, D., Jewcyszko, A., Lynch, A. M., & Urbaniak, C. (2022). Understanding the Complexities and Changes of the Astronaut Microbiome for Successful Long-Duration Space Missions. *Life*, 12(4), 1–47. <https://doi.org/10.3390/life12040495>

Voorhies, A. A., Mark Ott, C., Mehta, S., Pierson, D. L., Crucian, B. E., Feiveson, A., Oubre, C. M., Torralba, M., Moncera, K., Zhang, Y., Zurek, E., & Lorenzi, H. A. (2019). Study of the impact of long-duration space missions at the International Space Station on the astronaut microbiome. *Scientific Reports*, 9(1), 1–17. <https://doi.org/10.1038/s41598-019-46303-8>

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