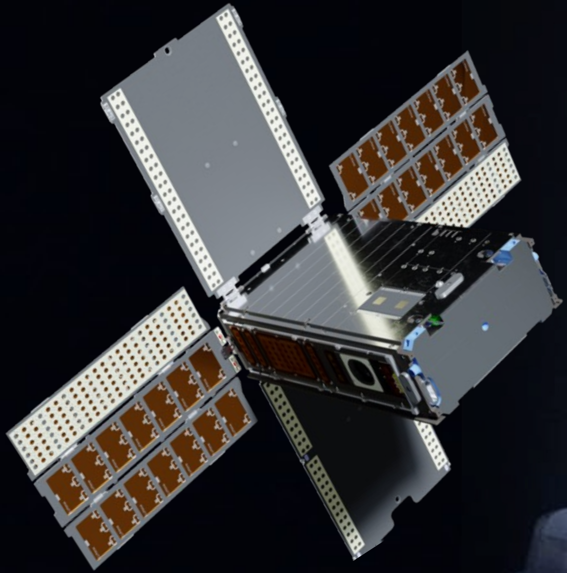
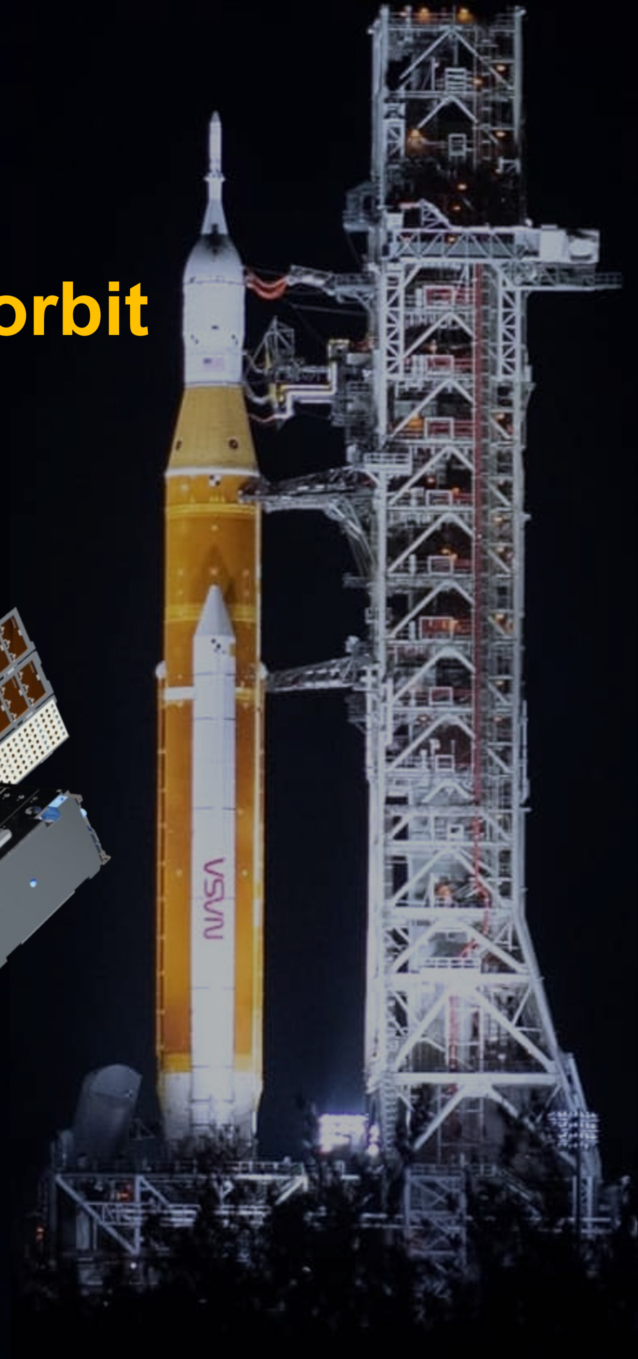
Space Biosciences
NASA AMES RESEARCH CENTER

Evolution of biological satellites: from low Earth orbit to NASA's BioSentinel deep space mission





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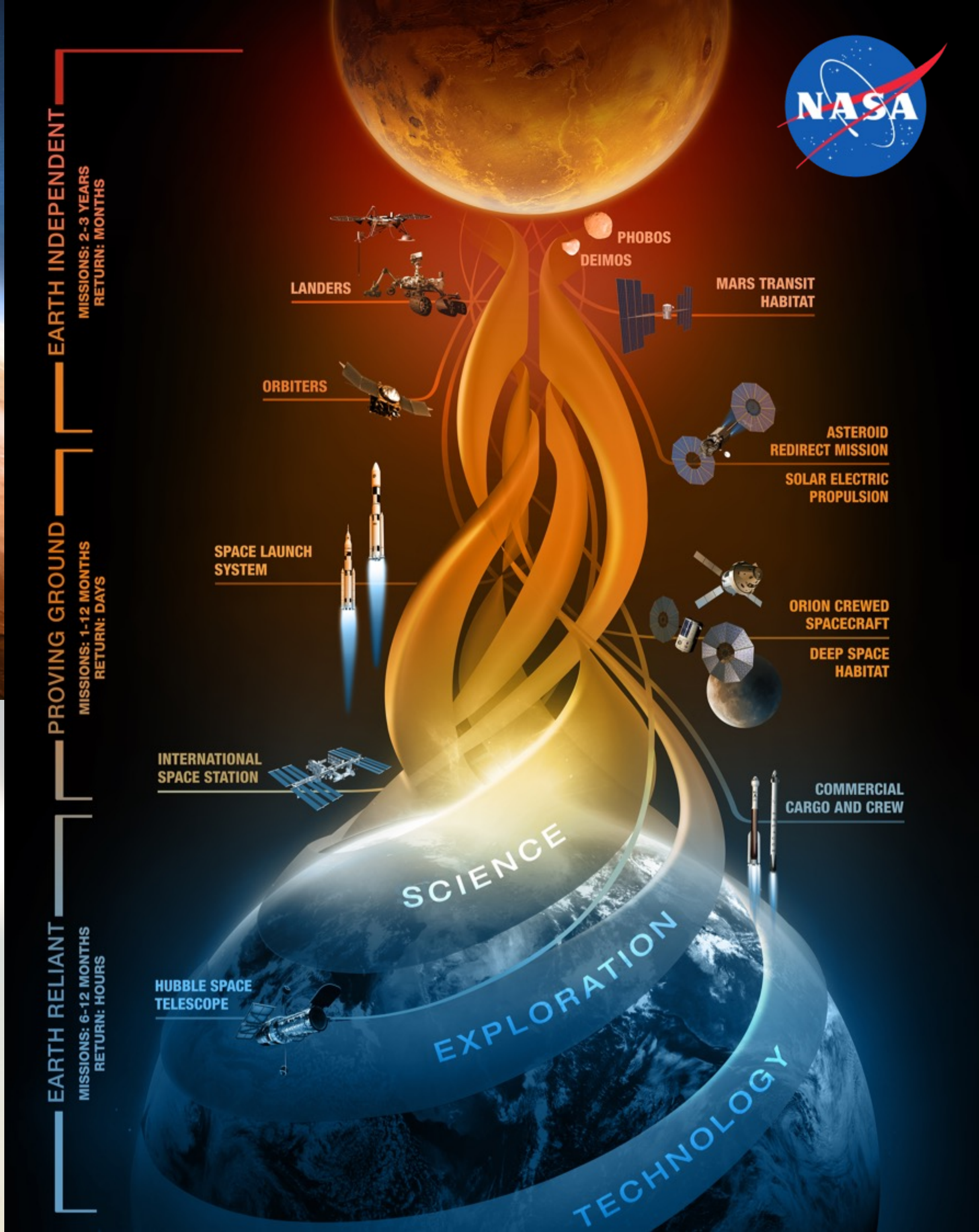
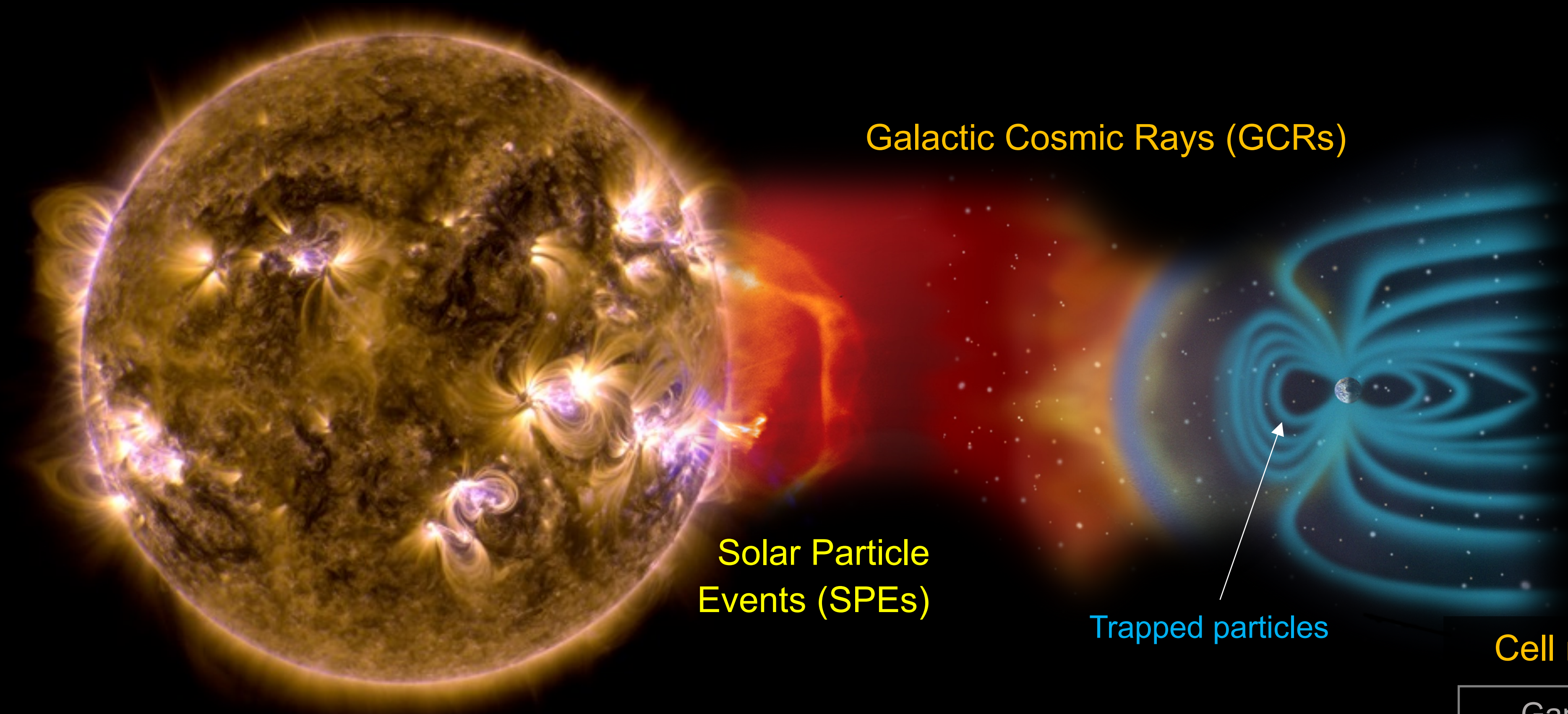
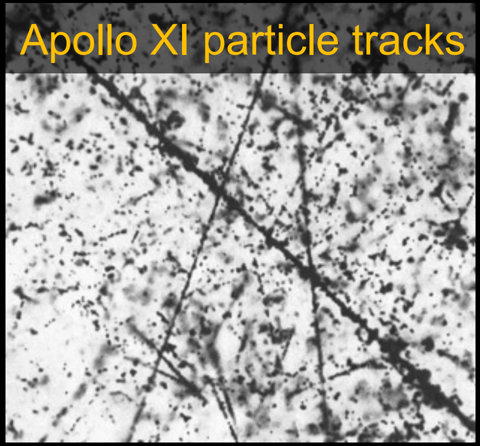
NASA'S JOURNEY TO
MARS


NASA
EARTH INDEPENDENT
MISSIONS: 2-3 YEARS
RETURN: MONTHS
PROVING GROUND
MISSIONS: 1-12 MONTHS
RETURN: DAYS
EARTH RELIANT
MISSIONS: 6-12 MONTHS
RETURN: HOURS
PHOBOS
DEIMOS
LANDERS
MARS TRANSIT HABITAT
ORBITERS
ASTEROID REDIRECT MISSION
SOLAR ELECTRIC PROPULSION
SPACE LAUNCH SYSTEM
ORION CREWED SPACECRAFT
DEEP SPACE HABITAT
INTERNATIONAL SPACE STATION
COMMERCIAL CARGO AND CREW
HUBBLE SPACE TELESCOPE
SCIENCE
EXPLORATION
TECHNOLOGY


NASA
EXPLORE
MOON to MARS

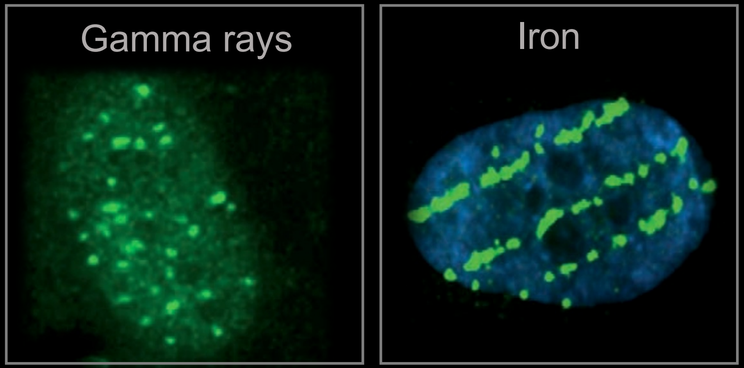
What are we going to encounter in deep space?





Schaefer *et al*. 1972

Cell nuclei showing DNA damage



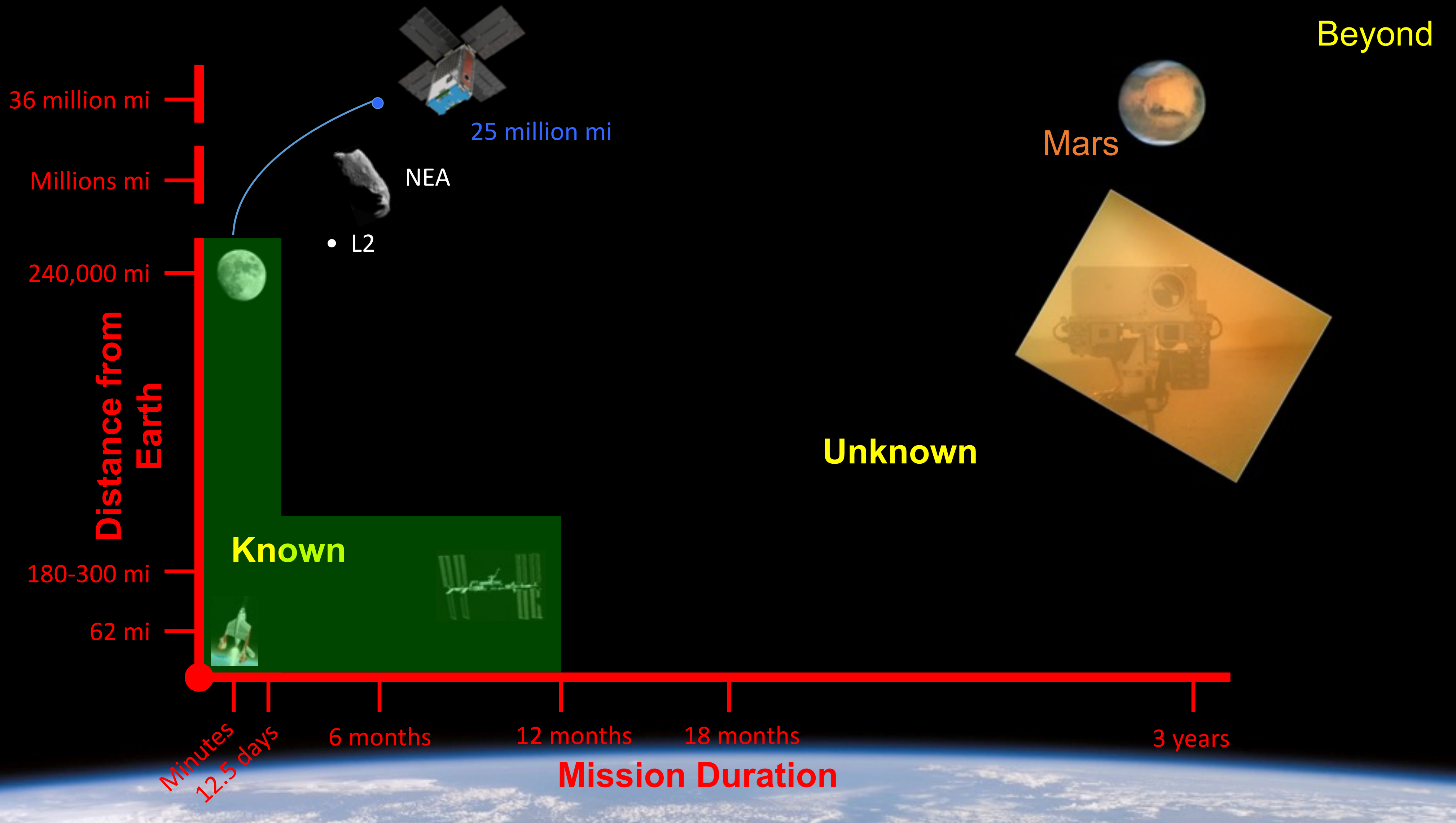
Cucinotta & Durante, 2006

- Both SPEs & GCRs are interplanetary, modulated by the 11-year solar cycle
- SPEs: sporadic, transient (mins to days); high proton flux (low-medium energy)
- GCRs: high-energy protons and highly charged, energetic heavy particles
- GCRs not effectively shielded; can break up into lighter, more penetrating pieces

<u>Challenges</u>: SPEs – unpredictable; large doses in short time

GCRs – biology effects poorly understood (but most hazardous)

The limits of life in space – as we know it – is 12.5 days on a lunar round trip or 1.2 years in LEO. As we send people further into space, we need to understand the biological risks and how they can be addressed

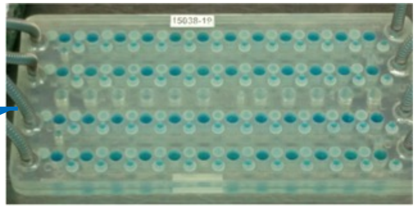


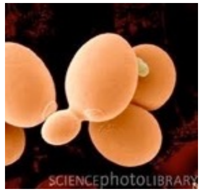
NASA Ames pioneering biological space missions

E. coli

GeneSat-1 (2006 / 3U): ***gene expression***

EcAMSat (2017 / 6U): ***antibiotic resistance***

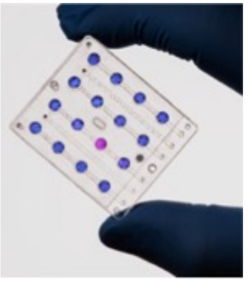




S. cerevisiae

PharmaSat (2009 / 3U): ***drug dose response***

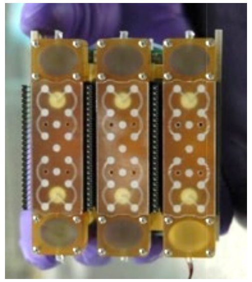
BioSentinel (2022 🥺 / 6U): ***cell damage repair***

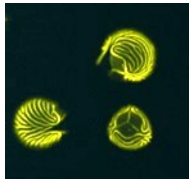


B. subtilis

O/OREOS* (2010 / 3U): ***survival, metabolism***

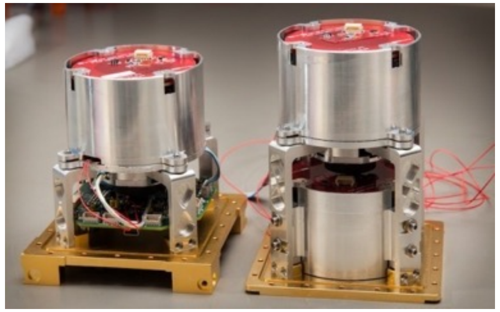
**Organism/Organic Response to Orbital Stress*





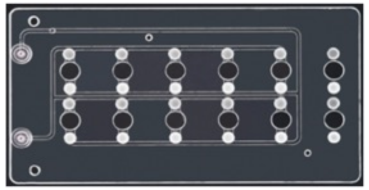
C. richardii

SporeSat-1 (2014 / 3U): **ion channel sensors, μ-centrifuges**

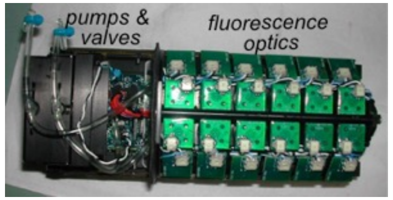


GeneSat mission: NASA's 1st CubeSat

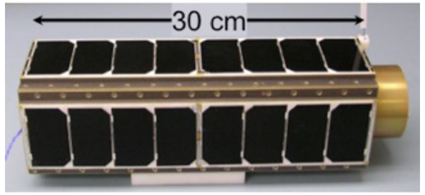
1st bio nanosatellite in Earth's orbit, 1st real-time, *in-situ* gene expression measurement in space

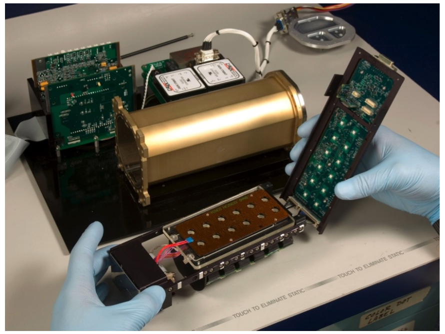


12-well fluidic card

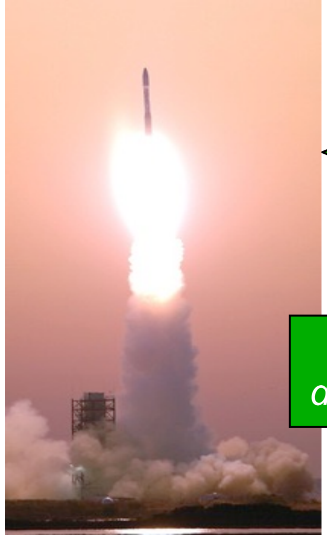


3U CubeSat



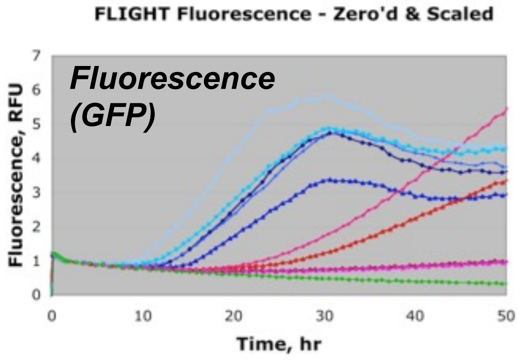


- Model organism: *E. coli* (~ 0.5 x 2 μm bacteria)
- Nutrient deprivation in dormant state (6 weeks)
- Launch: Dec 2006 to low Earth orbit (440 km)
- Nutrient solution feed upon orbit stabilization, grow *E. coli* in microgravity
- Monitor gene expression via GFP
- Monitor optical density: cell population

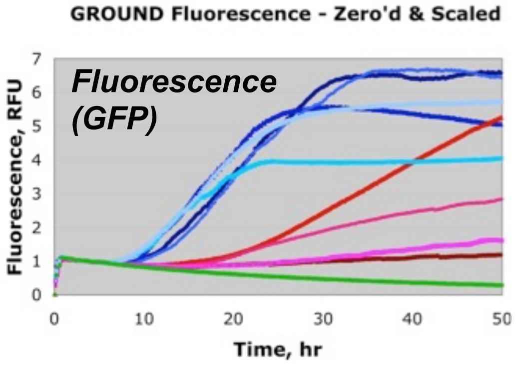


Dec 16, 2006

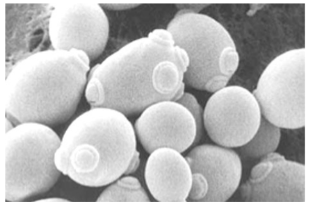
Telemeter data to Earth



Compare to ground data



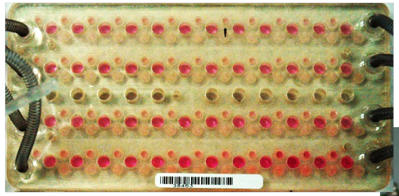
PharmaSat mission



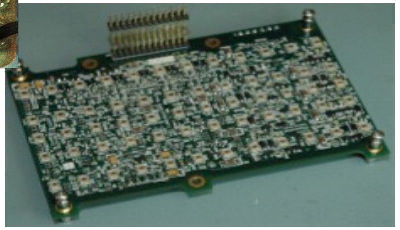
S. cerevisiae

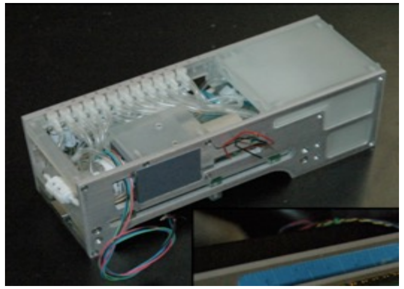
Effect of microgravity on yeast susceptibility to antifungal drug

- Launch: May 2009 to LEO (~450 km)
- Grow yeast in multiwell fluidics card in microgravity
- Measure inhibition of growth by antifungal
- Optical absorbance (turbidity: cell density)
- Metabolism indicator dye: alamarBlue (3-LED optical detection)
- Control + 3 concentrations of antifungal

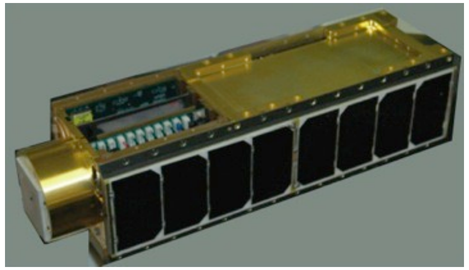


48-well fluidic card



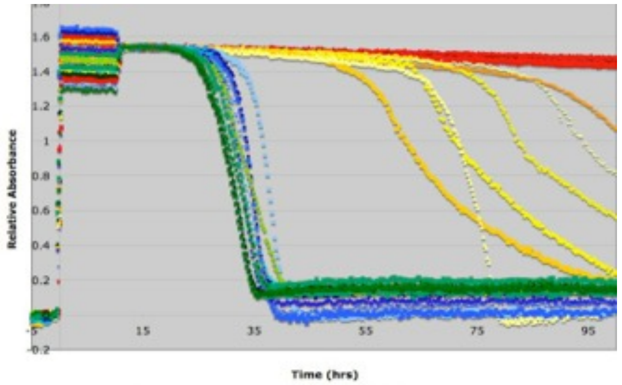


3U CubeSat

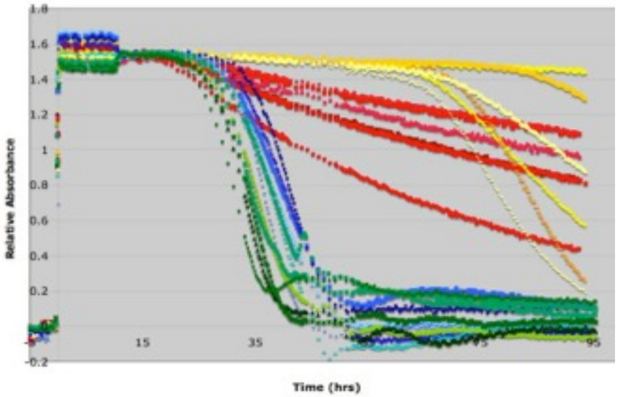


May 19, 2009

Ground



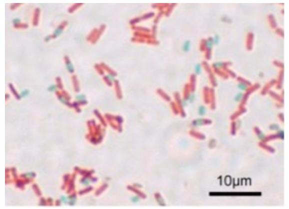
Spaceflight



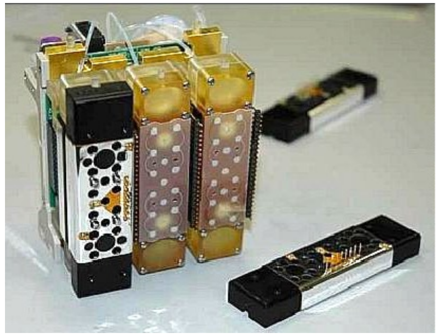
O/OREOS mission

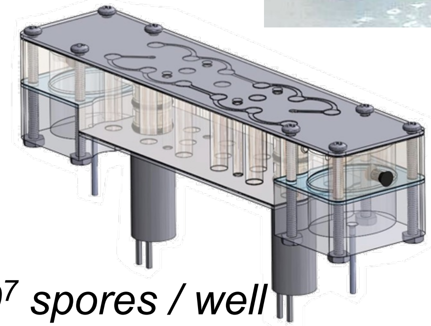
Organism / Organic Response to Orbital Stress (1st astrobiology CubeSat)

- Effects of space exposure on biological organisms (6 mo) & organic molecules (18 mo)
- Bus and 3U configuration derived from GeneSat & PharmaSat
- Launch: Nov 19, 2010 to highly-inclined orbit (~630 km)
- SESLO (Space Environment Survival of Living Organisms): monitor survival, growth, and metabolism of *B. subtilis* using *in-situ* optical density /colorimetry
- SEVO (Space Environment Viability of Organics): track changes in organic molecules and biomarkers: UV / visible / NIR spectroscopy

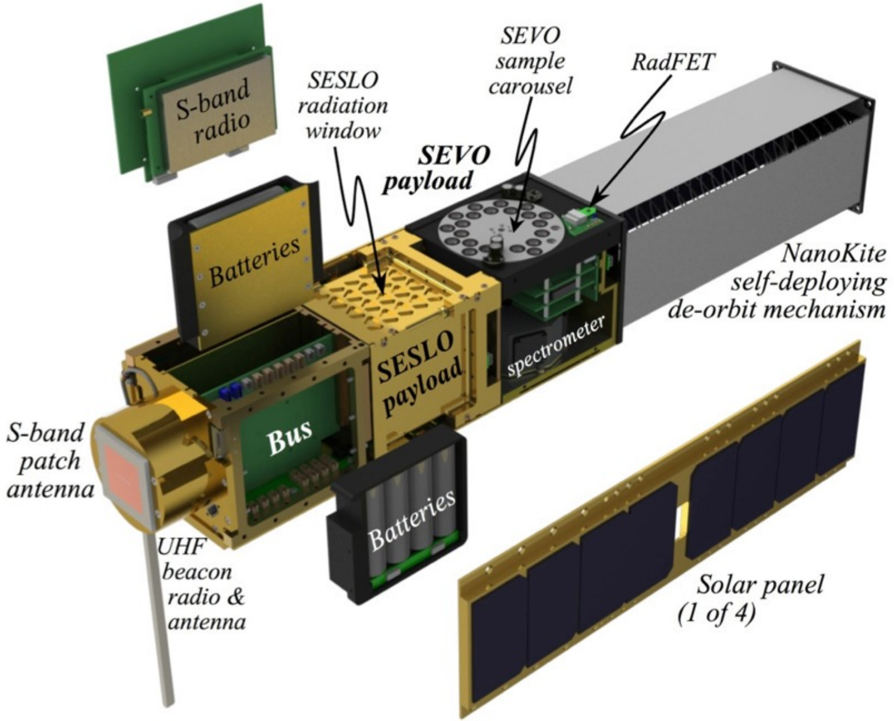


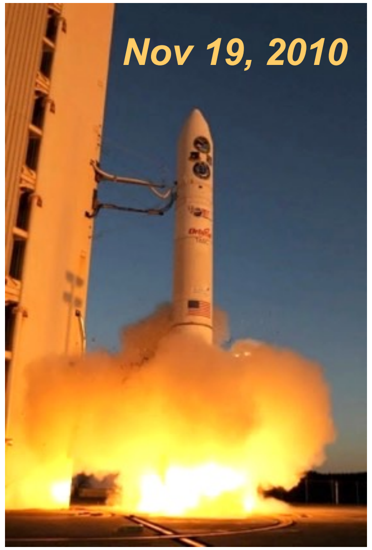
B. Subtilis
(dried spores)

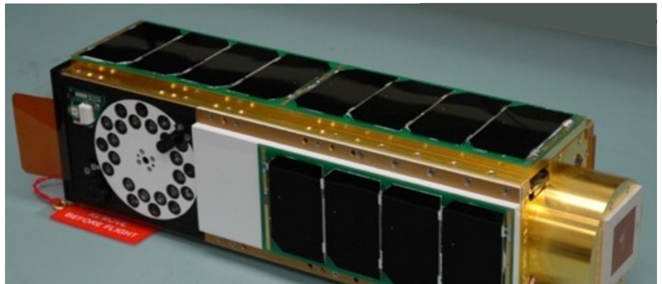




10^7 spores / well
(75 µL per well)





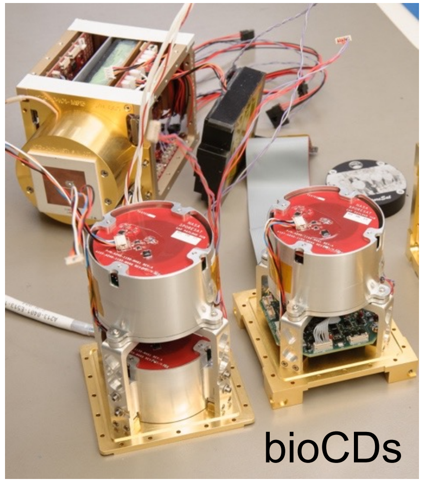


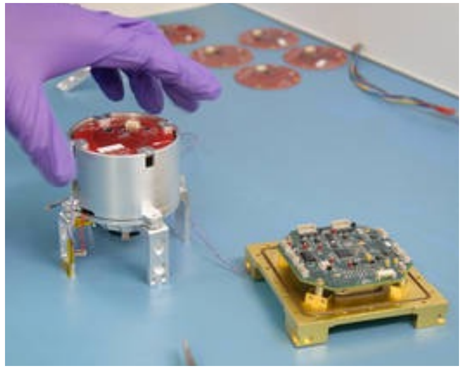
3U CubeSat

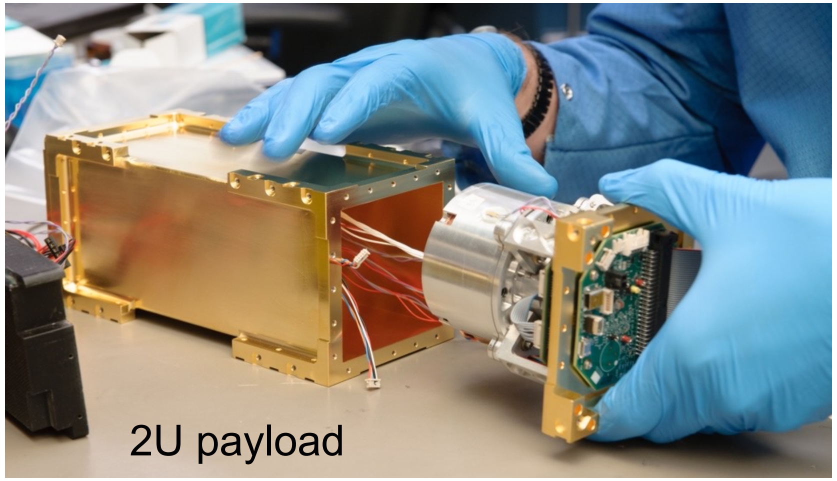
SporeSat mission

Gravitational response of fern spores via Ca^{2+} ion channel response

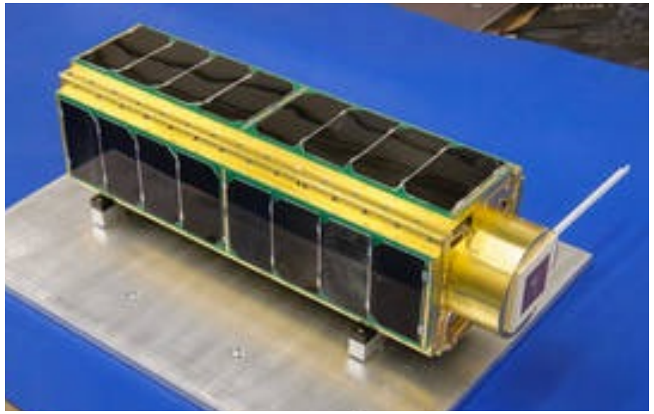
- Model organism: *Ceratopteris richardii* (aquatic fern spores)
- 2U payload (3U total)
- Launch: April 18, 2014 to LEO (325 km)
- Variable gravity in microgravity using 50-mm microcentrifuges
- 32 ion-specific [Ca^{2+}] electrode pairs (lab-on-a-chip devices, bioCDs)


bioCDs




2U payload


SpaceX CRS-3

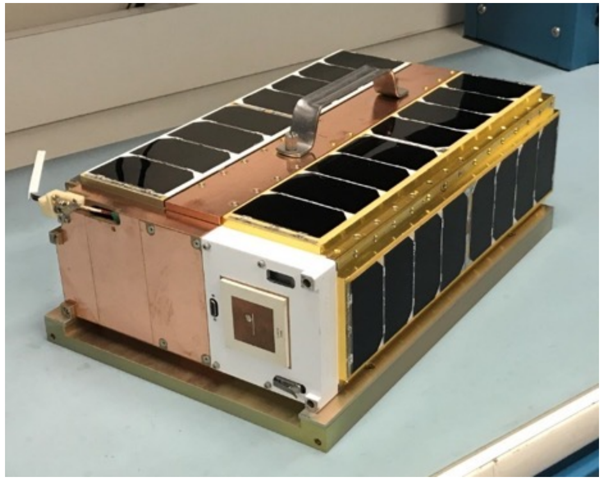

3U CubeSat

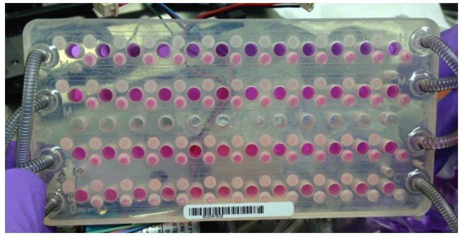
EcAMSat mission

E. coli AntiMicrobial Satellite mission (1st 6U bio CubeSat)

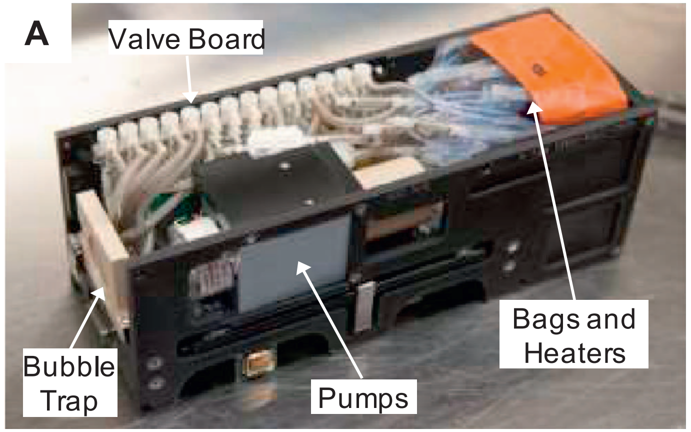
- Antibiotic resistance in microgravity vs. dose in uropathogenic *E. coli*
- Heavy reuse of PharmaSat design (6U format provided 50% more solar-panel power)
- Launch: Nov 12, 2017 (ISS deployment: Nov 20; ~400 km)
- 1st 6U bio satellite to be deployed from ISS

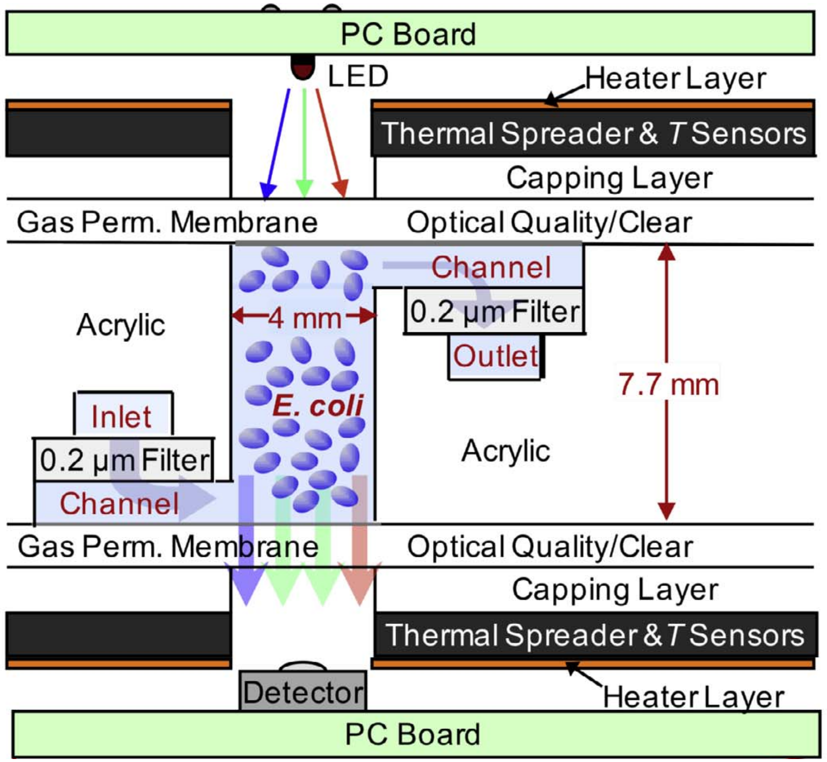
6U CubeSat



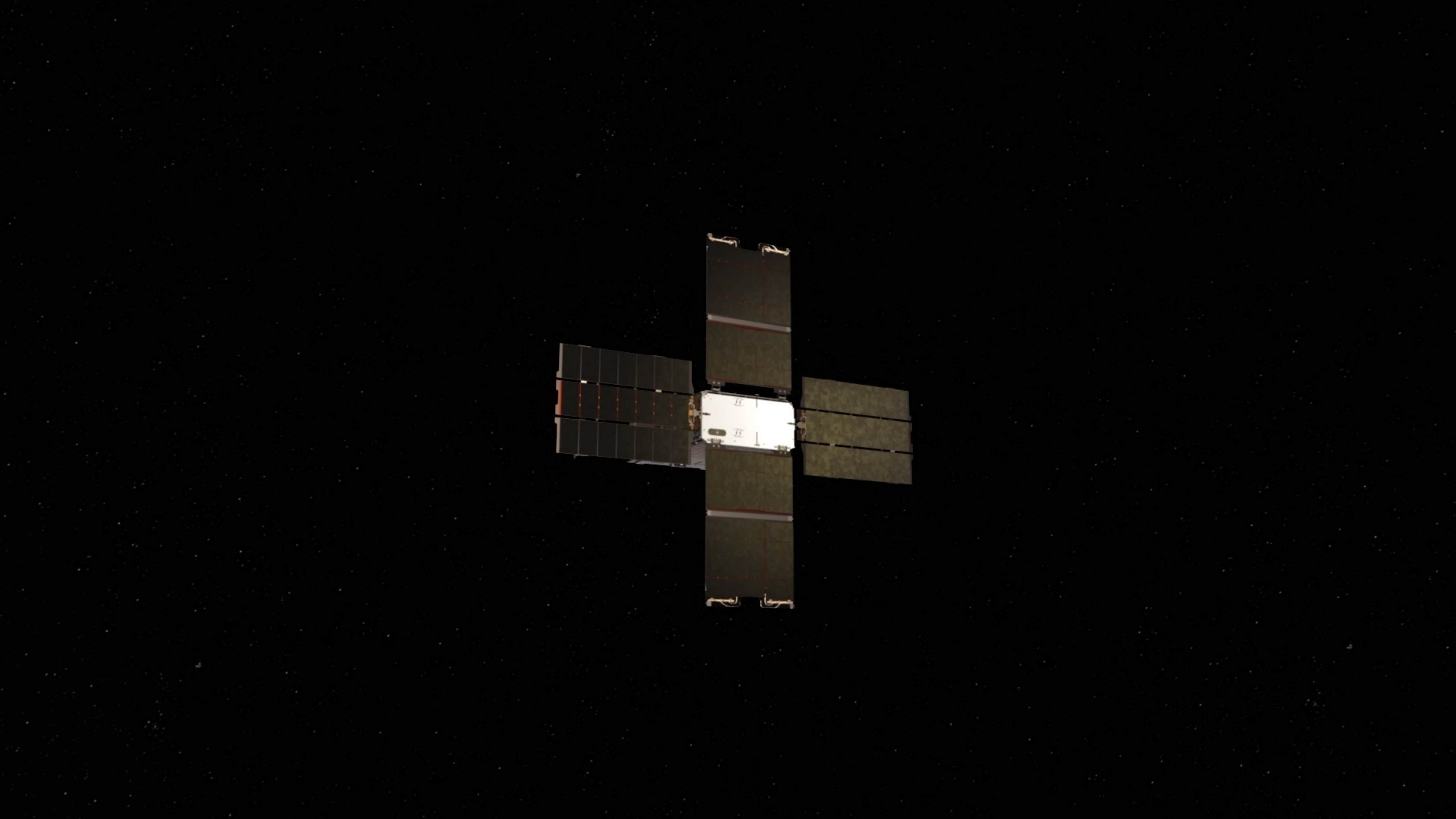


48-well fluidic card





Deployment from ISS



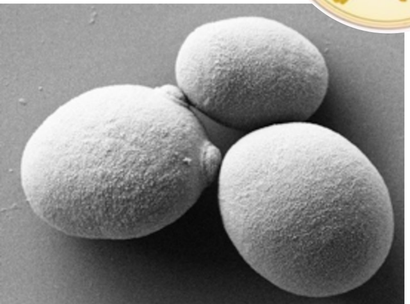
BioSentinel mission

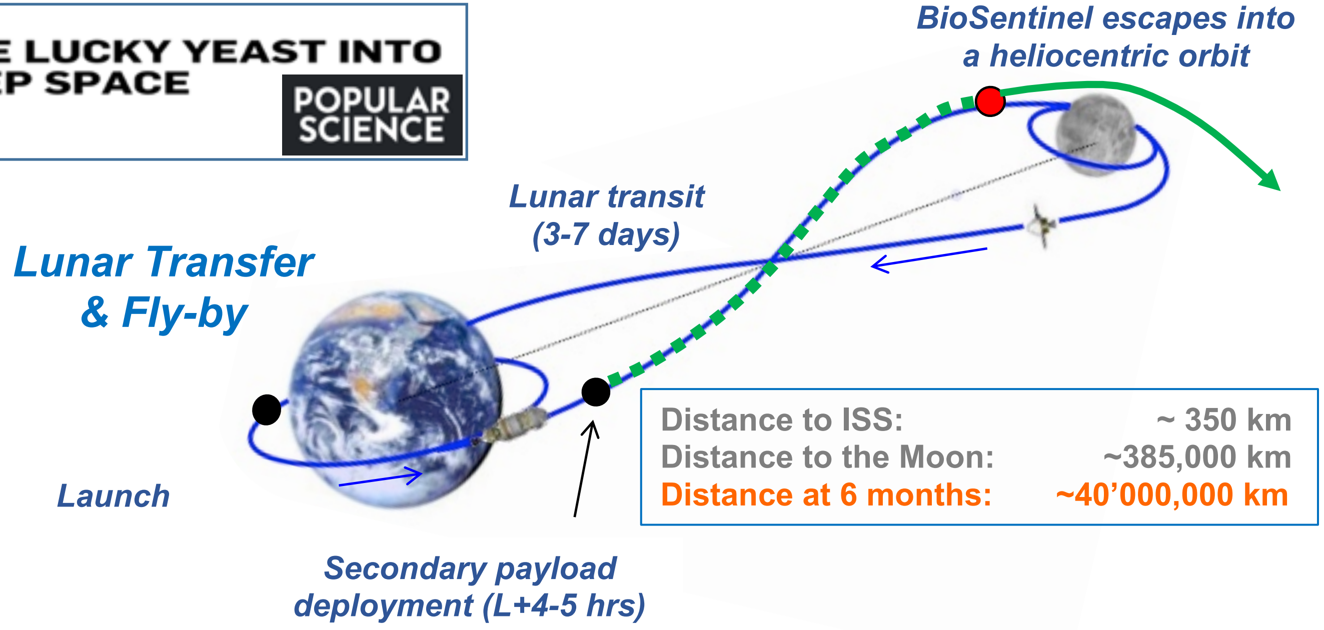
Objective: develop a deep space tool with autonomous life support technologies to study the biological effects of the space radiation environment

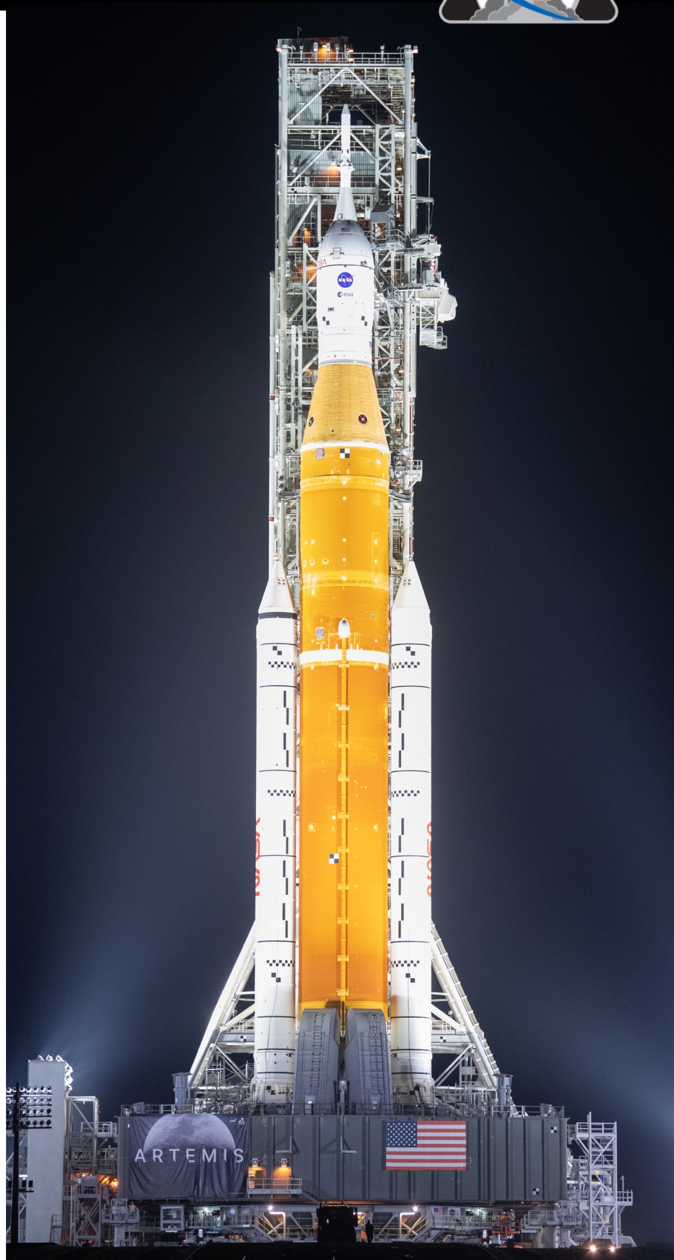
NASA's first biological study in interplanetary deep space

- First CubeSat to combine bio studies with autonomous capability & dosimetry beyond LEO
- Far beyond the protection of Earth's magnetosphere
- BioSentinel will allow to compare different radiation & gravitational environments (free space, ISS, lunar surface)

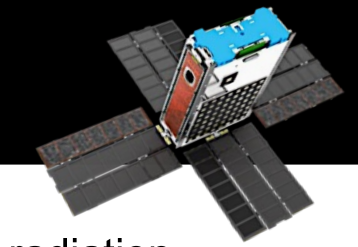
SPACE
NASA IS SENDING SOME LUCKY YEAST INTO RADIATION-FILLED DEEP SPACE
WHERE NO YEAST HAS GONE BEFORE
By Shannon Stirone May 15, 2015
POPULAR SCIENCE







What is BioSentinel?



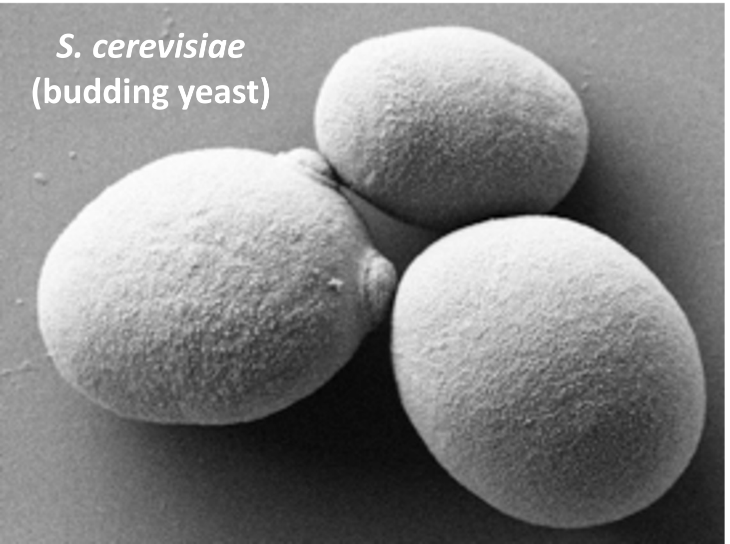
BioSentinel is a yeast radiation biosensor that will investigate the response to cell damage caused by space radiation, and will provide a tool to study the true biological effects of the space environment.

Why?

Space radiation environment's unique spectrum cannot be duplicated on Earth. It includes high-energy particles, is omnidirectional, continuous, and of low flux.

How?

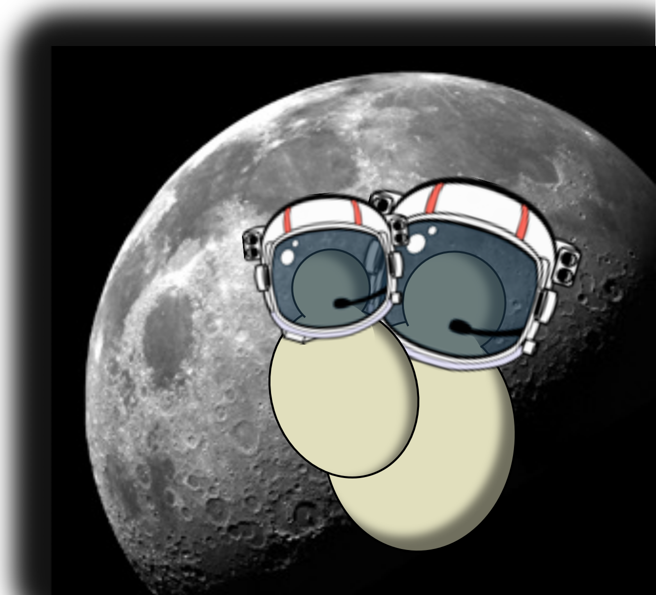
Lab-engineered *S. cerevisiae* cells will sense & repair direct (and indirect) damage. Yeast cells will remain dormant until rehydrated and grown using a microfluidic and optical detection system.



Why budding yeast?

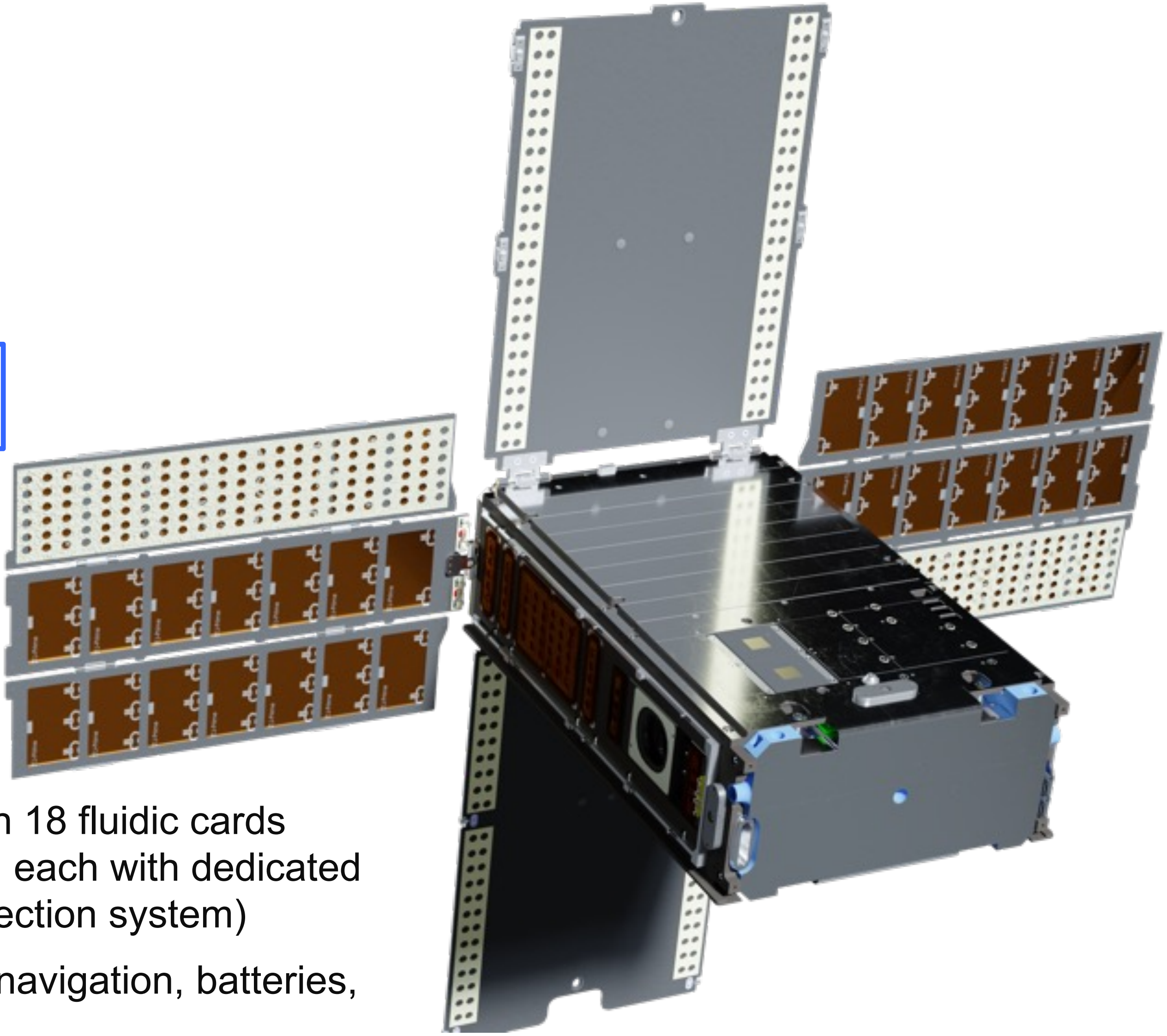
It is a eukaryote; easy genetic & physical manipulation; assay availability; flight heritage; ability to be stored in dormant state

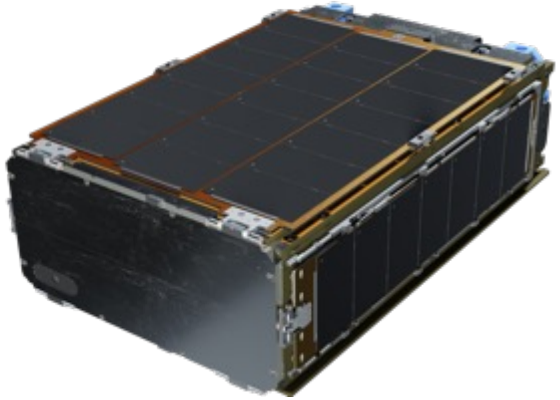
While it is a simple model organism, yeast cells are the best for the job given the limitations & constraints of spaceflight



BioSentinel: a bio CubeSat for deep space

6U Cubesat
37 x 24 x 12 cm ~ 10 L

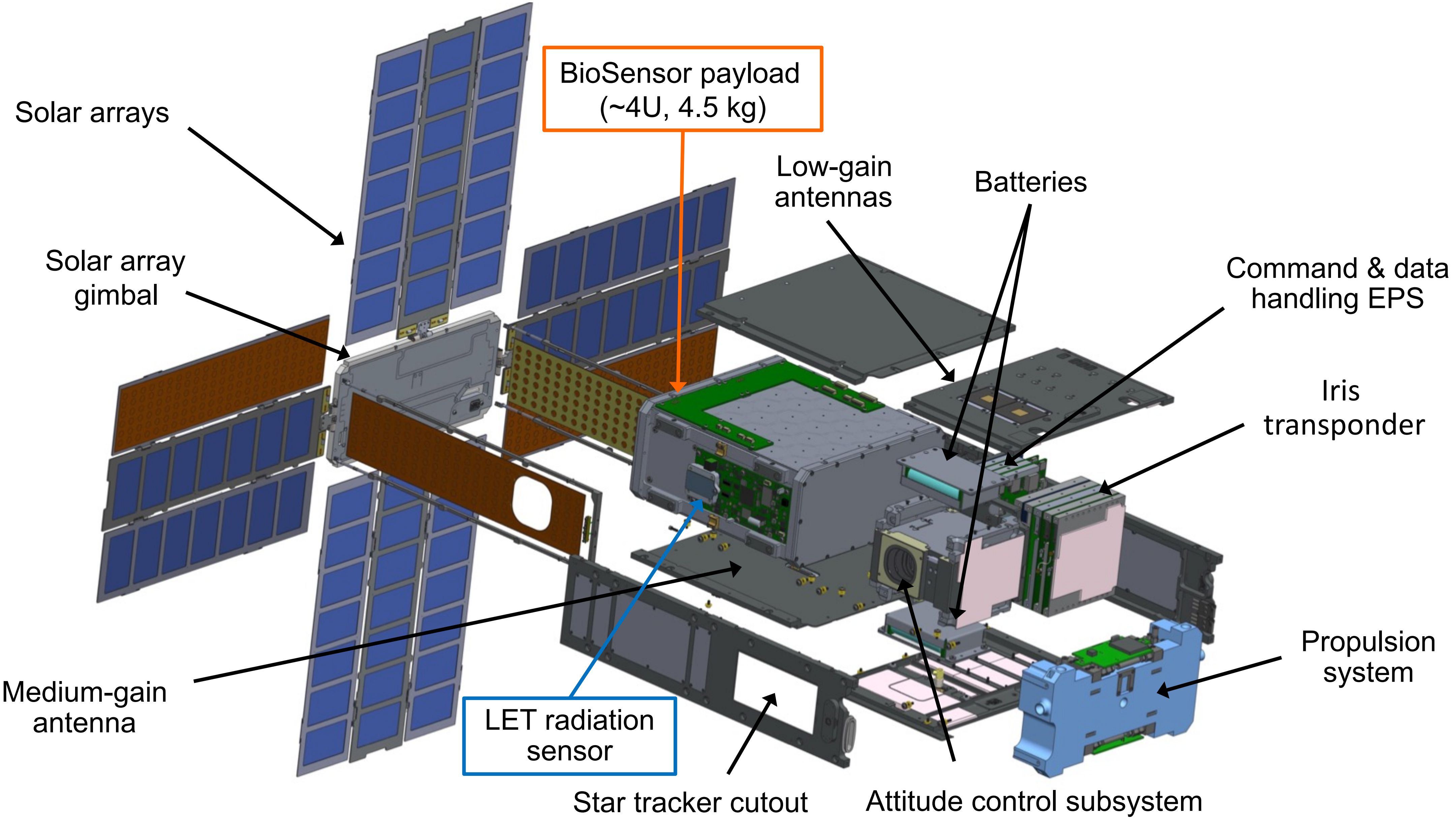




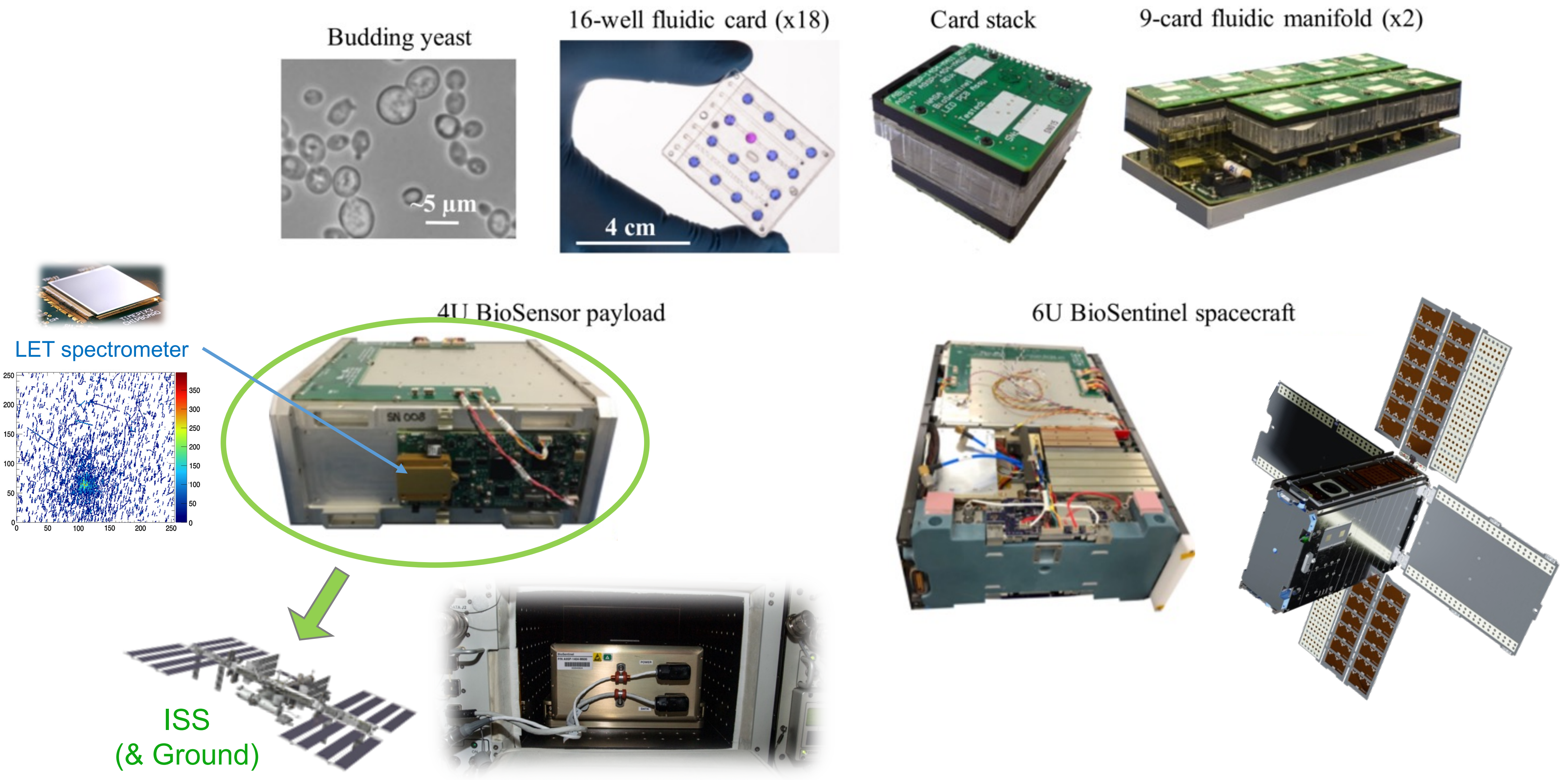
Spacecraft sections:

- BioSensor payload (~4U with 18 fluidic cards loaded with desiccated cells, each with dedicated thermal control & optical detection system)
- Spacecraft bus (propulsion, navigation, batteries, transponder, star tracker…)

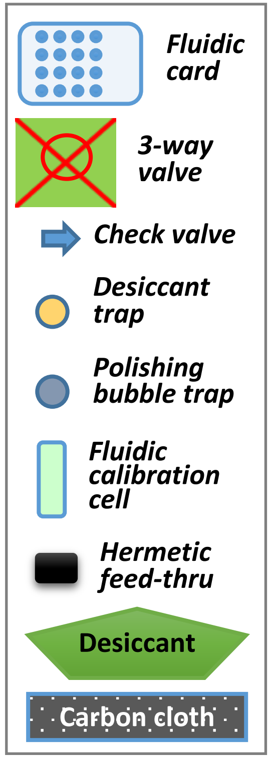
BioSentinel: a bio CubeSat for deep space

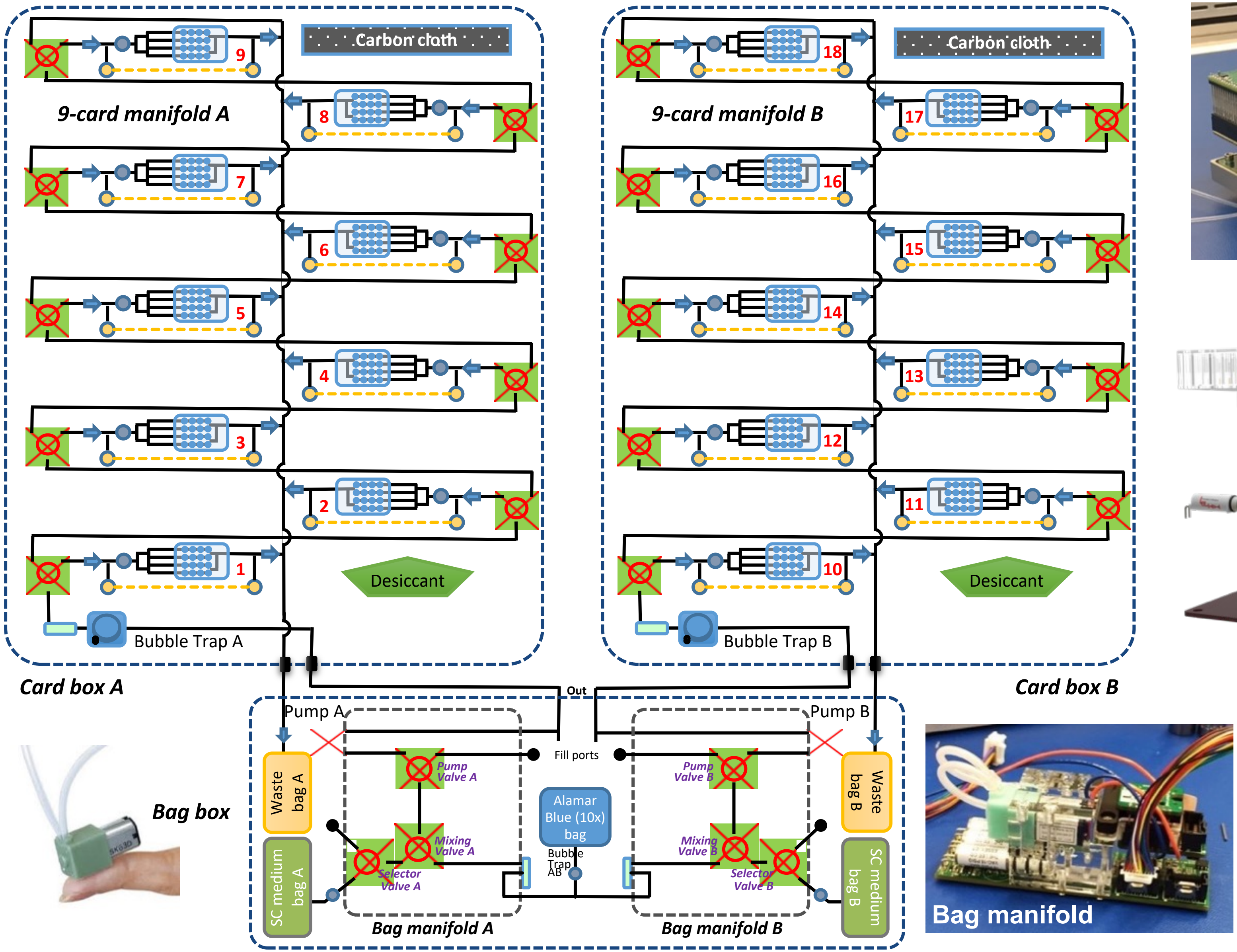


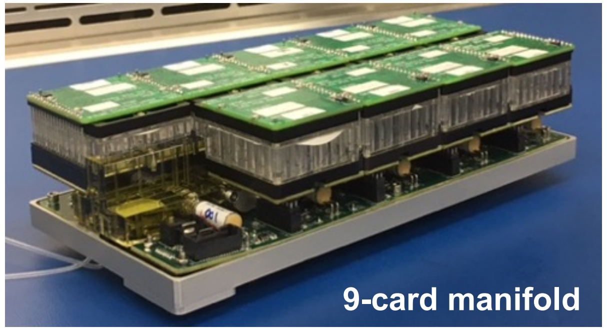
BioSentinel: a bio CubeSat for deep space

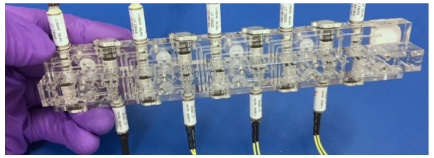


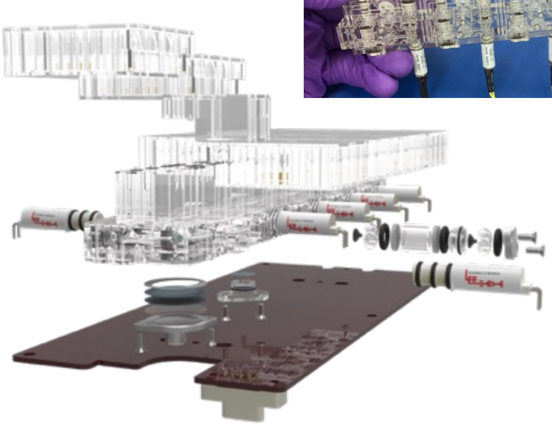
BioSentinel: microfluidic system









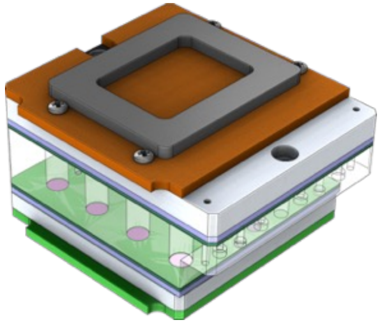


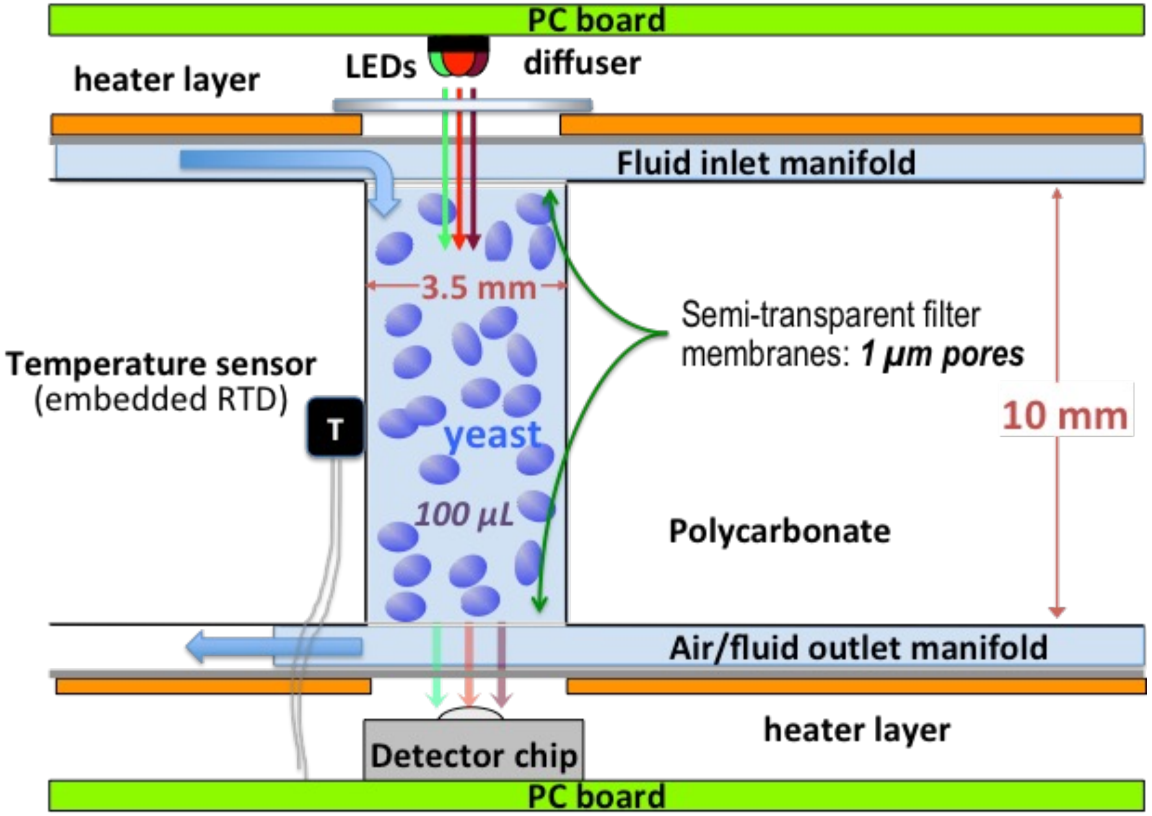
Manifold-integrated components:

- *2 pumps*
- *24 active & 38 check valves*
- *2 main bubble traps*
- *4 optical calibration cells*
- *18 fluidic cards with small bubble traps & desiccant traps*
- *288 microwells total*

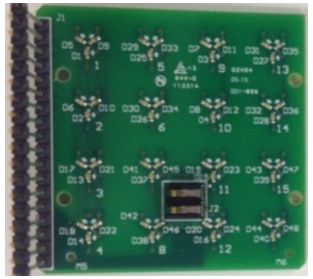
BioSentinel: microfluidic card

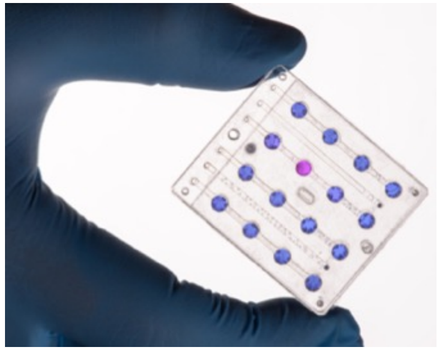
Microfluidic card (x18)



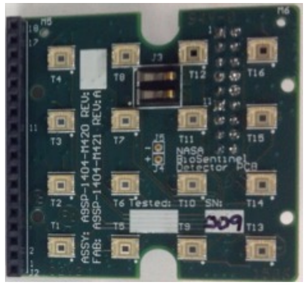


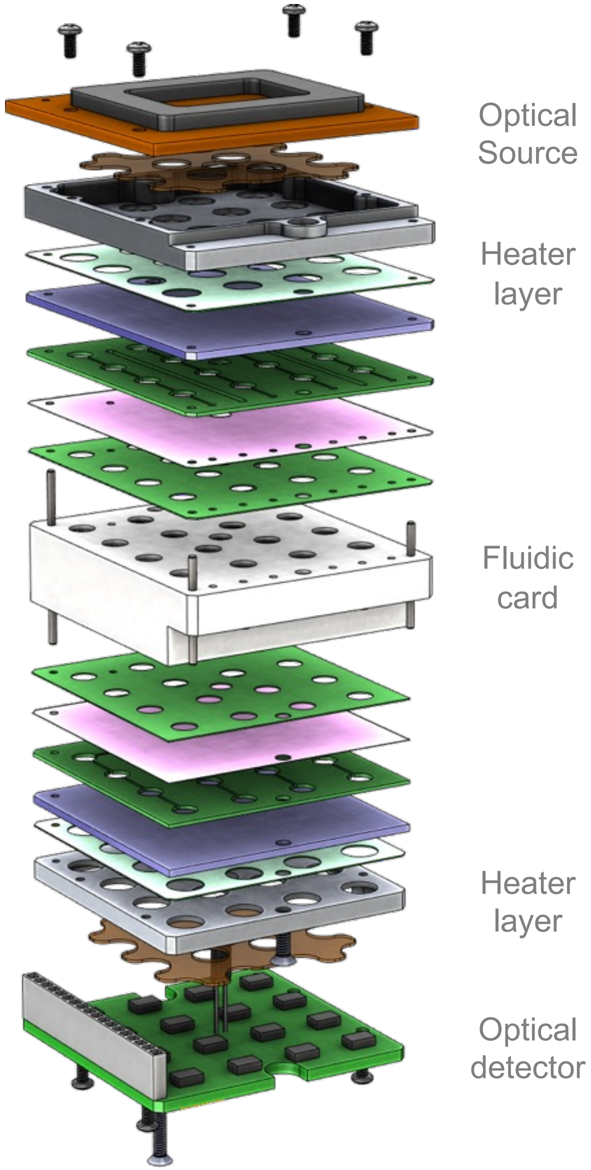
3-LED emitter





Photodiode detector array





BioSentinel: optical detection system

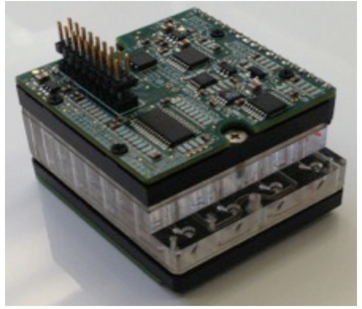
Dedicated 3-color optical system at each well to track growth *via* optical density and cell metabolic activity *via* dye color changes

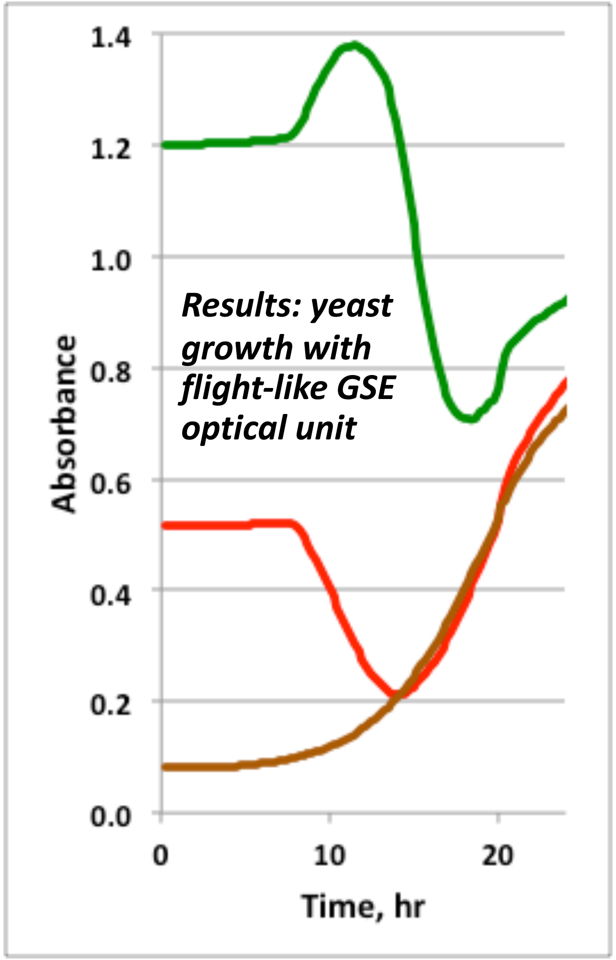
LEDs:

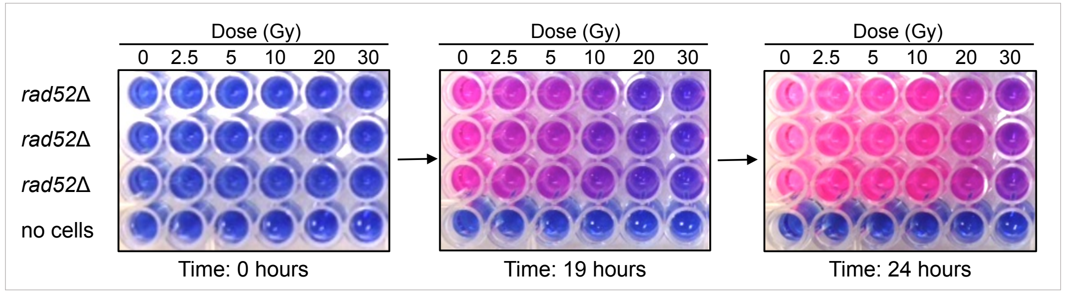
570 nm (green, measures pink)

630 nm (red, measures blue)

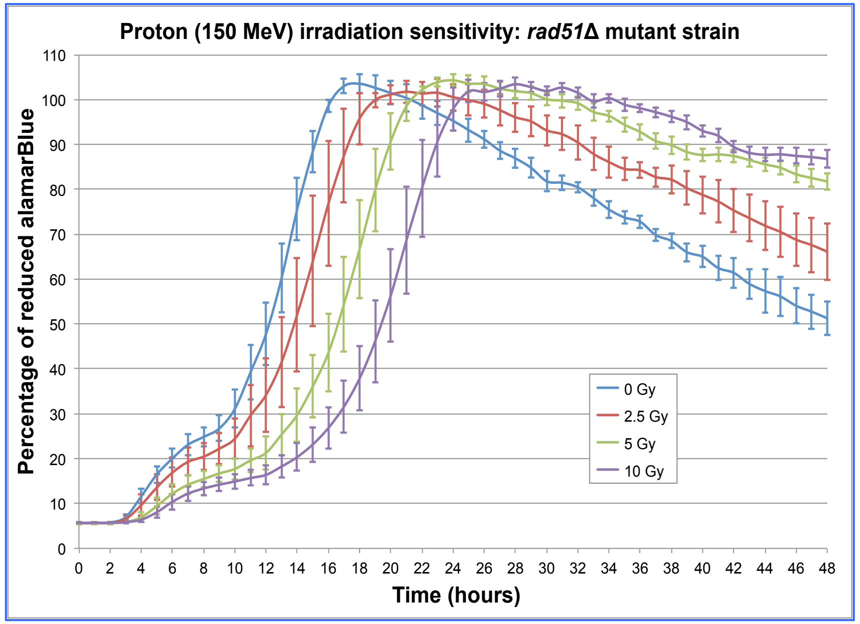
850 nm (infrared, measures growth)





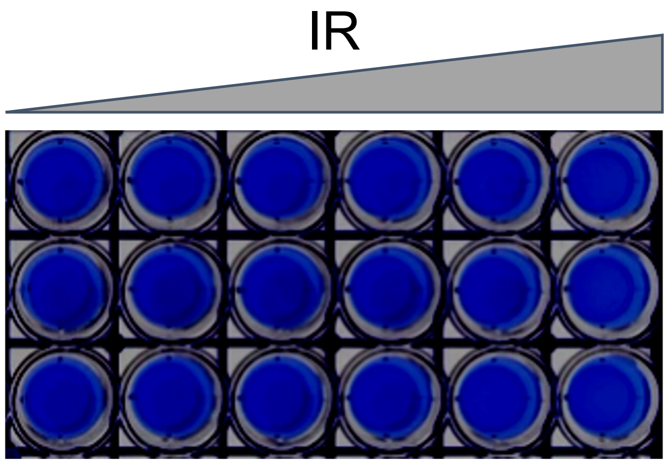


alamarBlue turns pink when cells are metabolically active

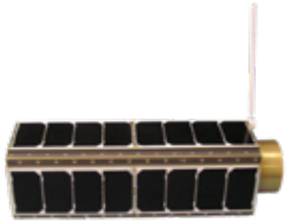


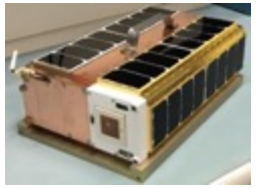
HR repair defective cells show sensitivity to ionizing radiation

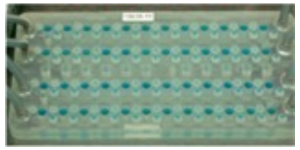
BioSentinel: how did we get here?



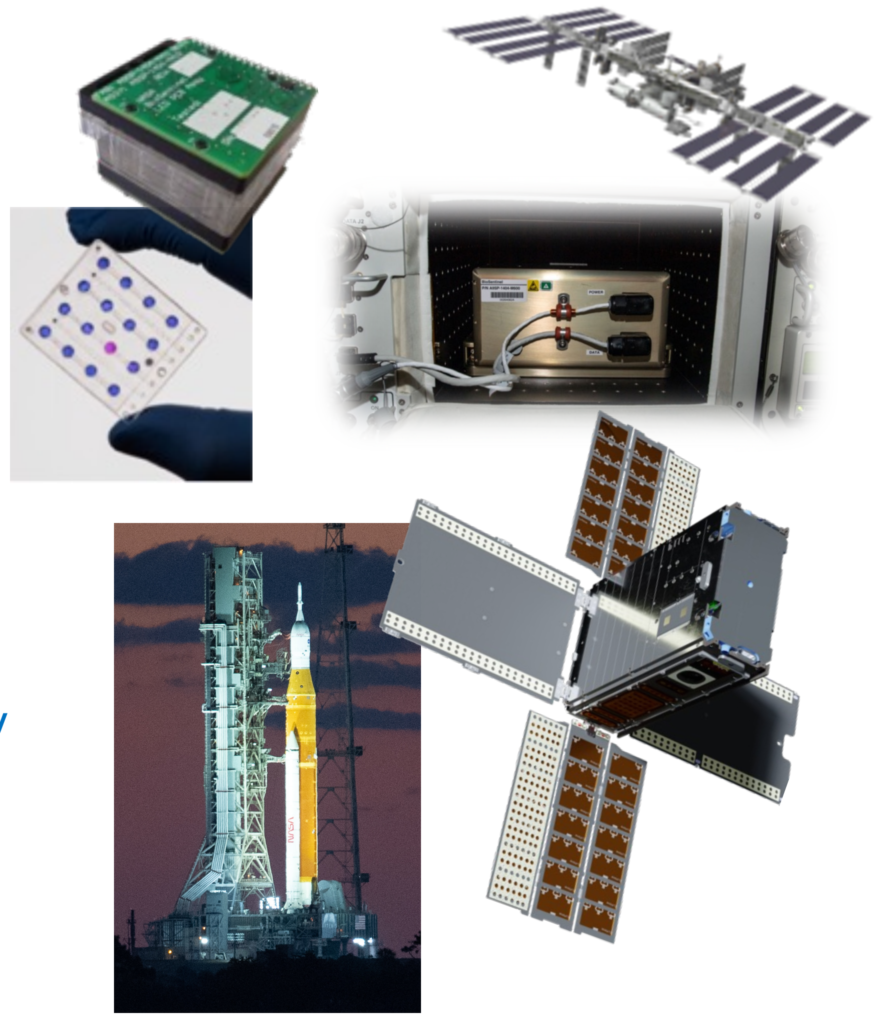
Instrument & flight heritage



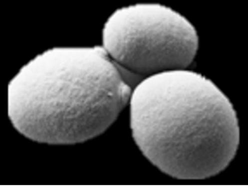




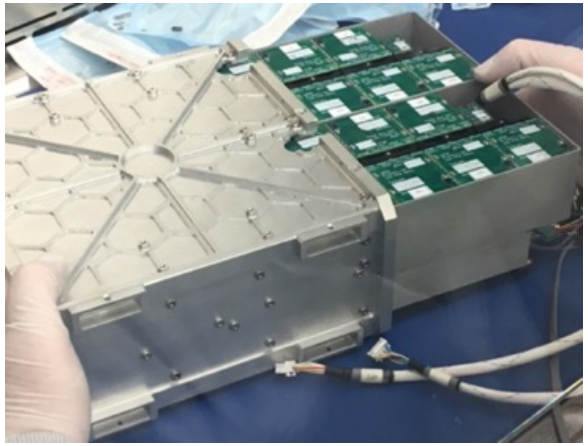
- Model organism & strain selection
- How to fly the biology? Dry/wet?
- Long-term viability & biocompatibility
- Materials, sterilization method, reagents, fluidic components…
- Multiple fluidic cards; 288 wells
- LEO control (and ground)
- Data telemetry & processing



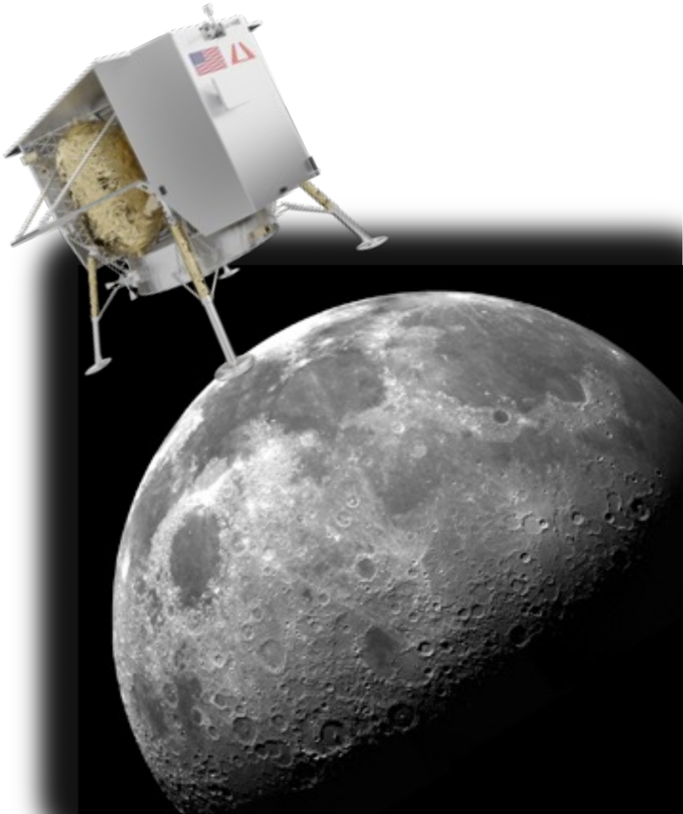
BioSentinel: lessons learned & LEIA



LEIA: Lunar Explorer Instrument for space biology Applications

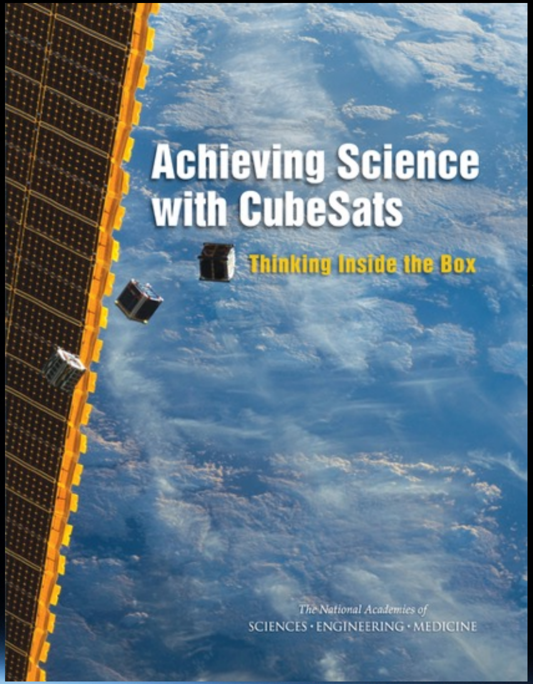


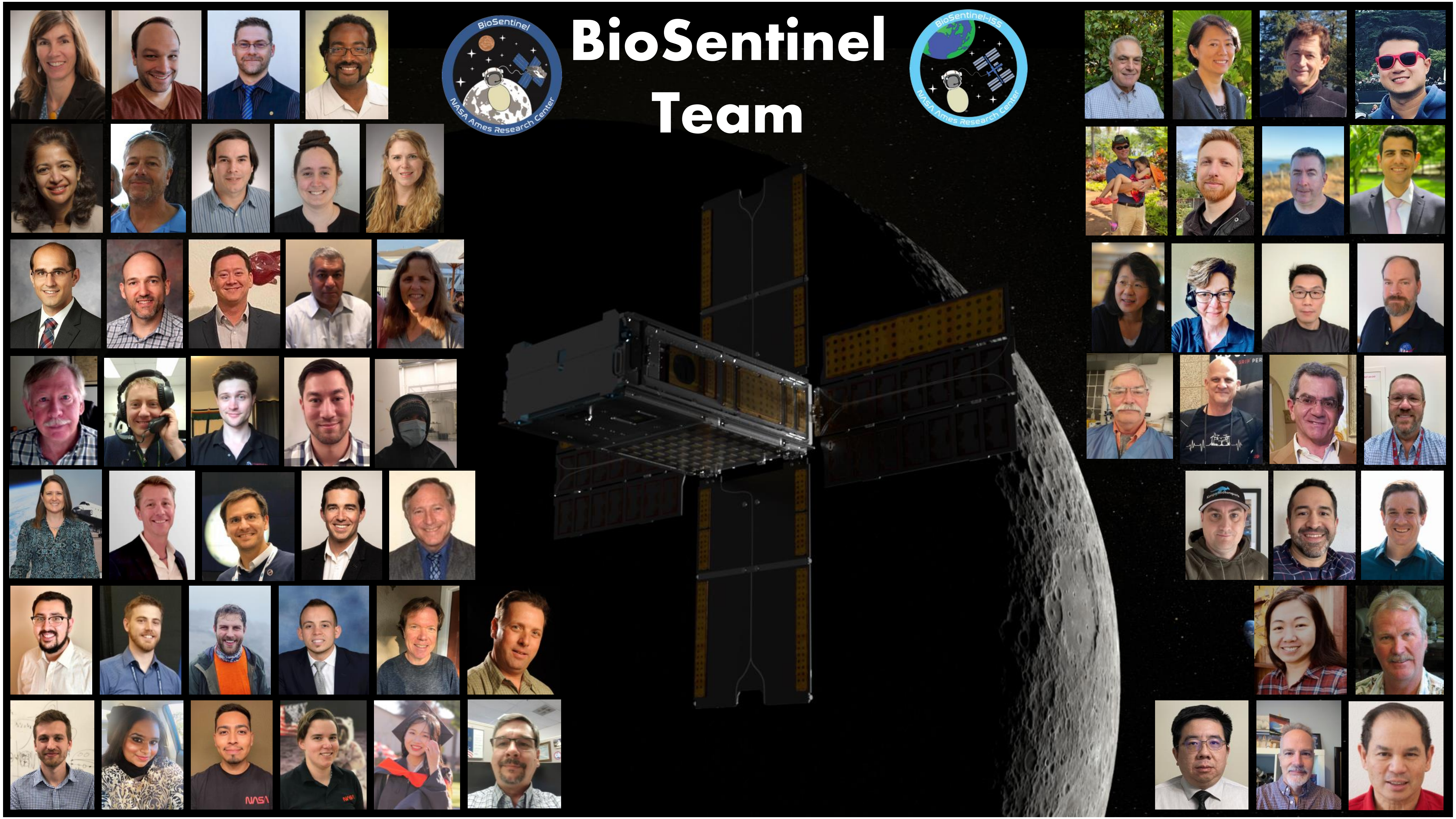
- Three awarded Space Biology ground investigations (two ARC PIs)
- Significant instrument heritage from BioSentinel
- ARC team selected for a PRISM2 award to perform science experiments on the lunar surface (synthetic biology & stress/damage response)
- LEIA will have additional countermeasures to improve long-term viability (*e.g.*, late load of biology and improved desiccant storage)
- LEIA suite also include a LET spectrometer and a fast neutron detector
- Power & data from CLPS lander



Why CubeSats?

- Small satellites are very capable: technology miniaturization & maturation
- Access to space: multiple low-cost launches possible (test → learn → iterate)
- Excellent education vehicle (worldwide)
- Autonomous operations
- Technology migration: ISS; free-flyers; landers/orbiters for moon, Mars & other planets

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Thank you!