

ELM: Europa Luminescence Microscope

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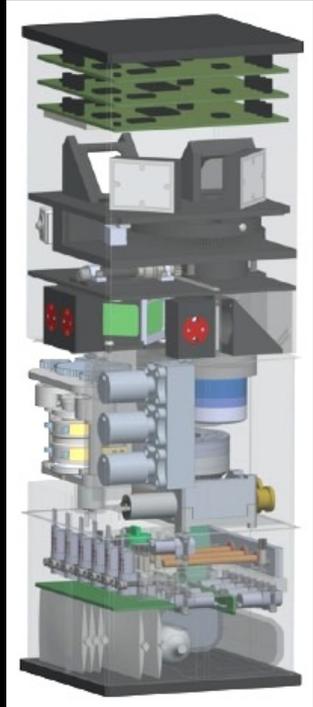
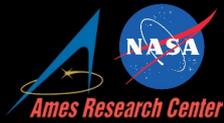
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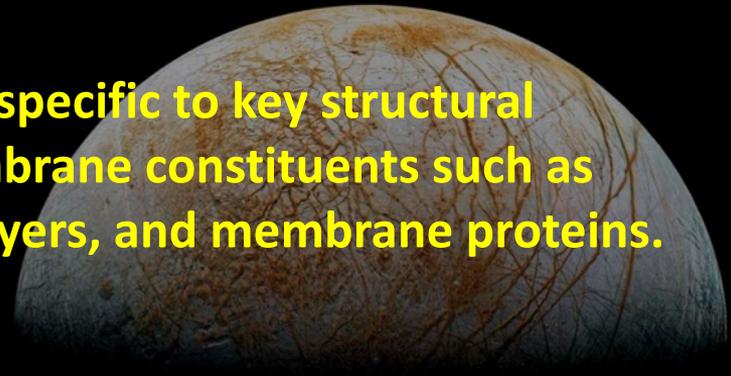
ELM: Europa Luminescence Microscope

(Micro)fluidics-based fluorescence and bright field imaging microscope with sub-micron resolution

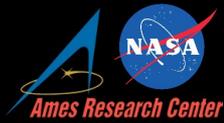


3 Operational Modes:

- 1) **Bright field imaging for visual characterization and context.**
- 2) **Using DUV and visible light-emitting diodes (LEDs) for the excitation of native luminescence in the samples; mineralogical and biological.**
- 3) **Utilizing fluorescence stains specific to key structural biomarkers, i.e., typical membrane constituents such as fatty acids, phospholipid bilayers, and membrane proteins.**



ELM Science Objectives



Designing to broadly and specifically address the Europa Lander Study Report

Report Defined Goals:

- Search for evidence of life on Europa (redefined as biosignatures)
- Assess the habitability of Europa



Objectives:

1) *Identify and characterize morphological, textural, indicators*

Sub-micron bright field imaging; Spatial Fluorescence

2) *Detect and characterize inorganic indicators*

Native Fluorescence - mineralogy

3) *Detect and characterize any organic indicators*

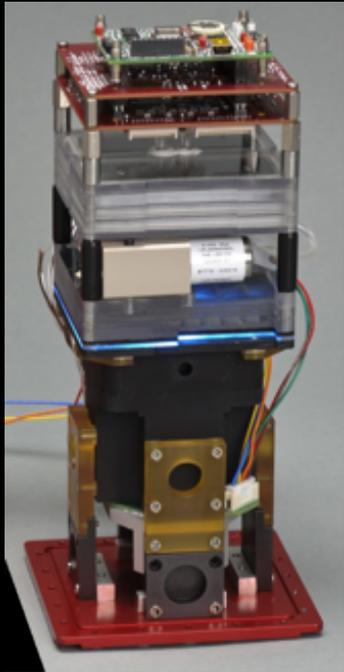
Proteins, polypeptides (i.e. amine-containing compounds), Lipids in organized structures (e.g. membranes), Nucleic Acids

4) *Determine the presence of environmental factors essential for life (habitability)*

Embedded sensors (pH, Eh, Conductivity)

ELM Technology Heritage

Fluorescence Analysis for In-situ Research on Nanosatellites (FLAIR) Imager
Part of the Microsatellite in-Situ Technologies (MisST) project



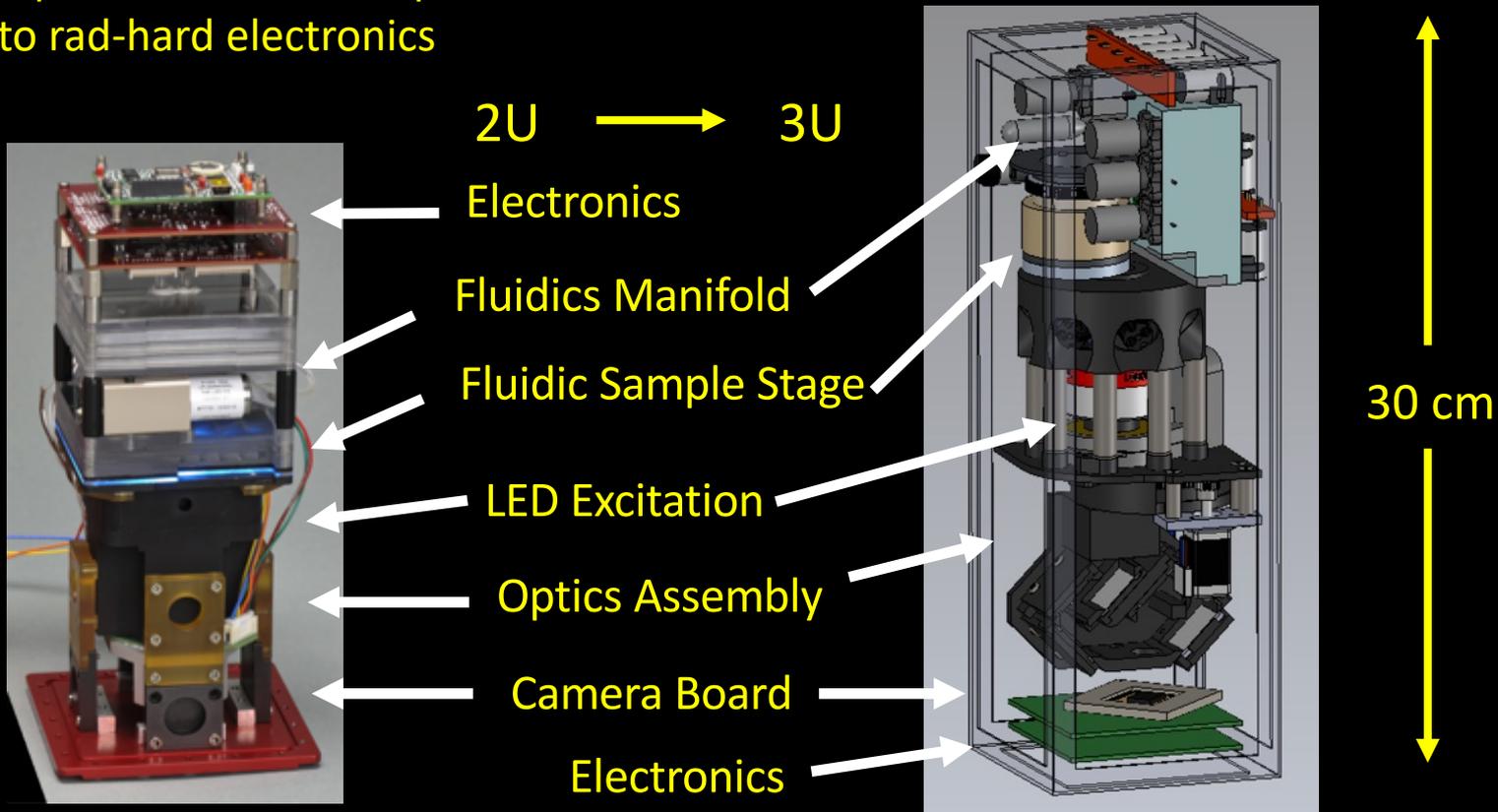
- 2U (10 cm x 20 cm) CubeSat payload
- Designed for biological investigations in low-earth orbit
- Autonomous microfluidics and imaging
- Dual-wavelength LED fluorescence excitation (blue & amber)
- Dual-bandpass emission filter
- White LED dark field imaging (<math><4 \mu\text{m}</math> resolution with LEO qualified imager; 2 $\mu\text{m}</math> resolution w/upgraded imager)$
- Pressure, Temperature, RH Sensors

COLDTech and ICCE2: Advanced microfluidic processor for filter-capture and fluorescence labeling. Submicron resolution UV-Vis fluorescence imaging optical system

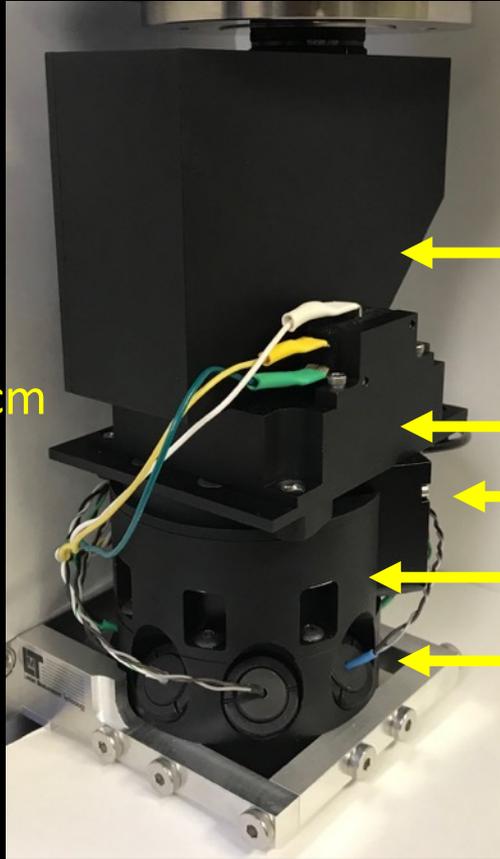
TRL migration LEO → Europa

Primary Growth Drivers:

- Submicron spatial resolution requirement
- Migration to rad-hard electronics



ELM Microscope



Folded Optics

Filter Wheel

Piezo Stage

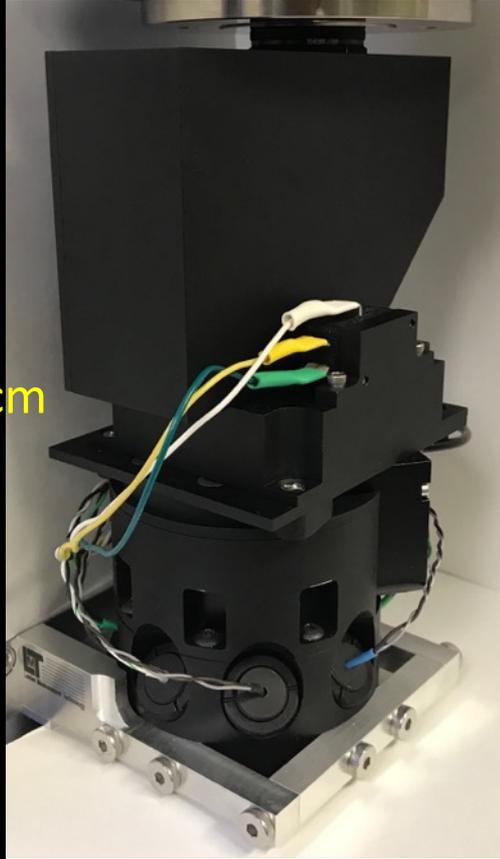
Objective

LED Excitation

Field-of-View	761 x 761 μm
Spatial Resolution	< 0.8 μm
Piezo Z-motion	up to 400 μm
Piezo Positioning	1 nm accuracy
LEDs 1 & 5	275 nm
LEDs 2 & 6	375 nm
LEDs 3 & 7	470 nm
LEDs 4 & 8	525 nm
Emission Filters	334, 470, 529, 579

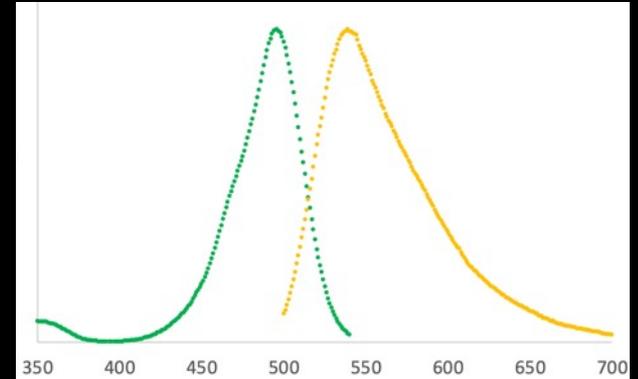
Components have passed radiation testing to 300 krad, including: bandpass filters (single- and multi-band); LED modules; lenses and lens materials.

ELM Microscope

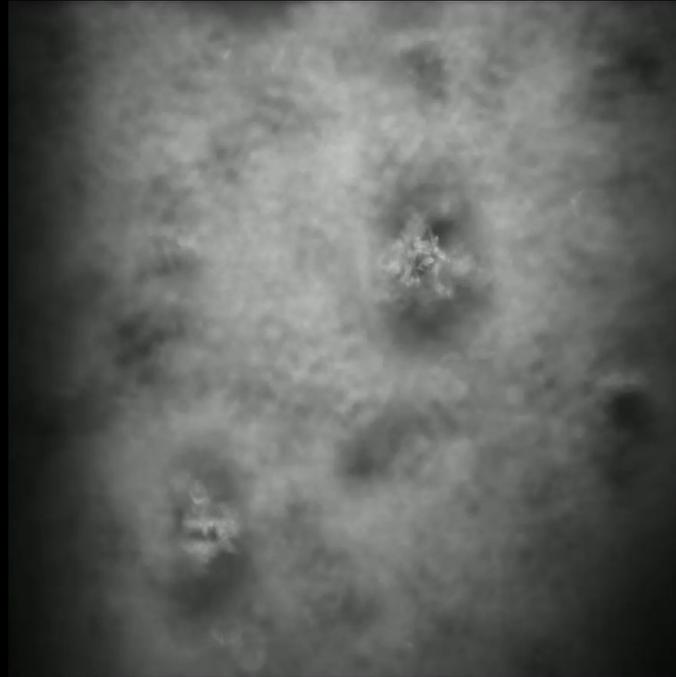
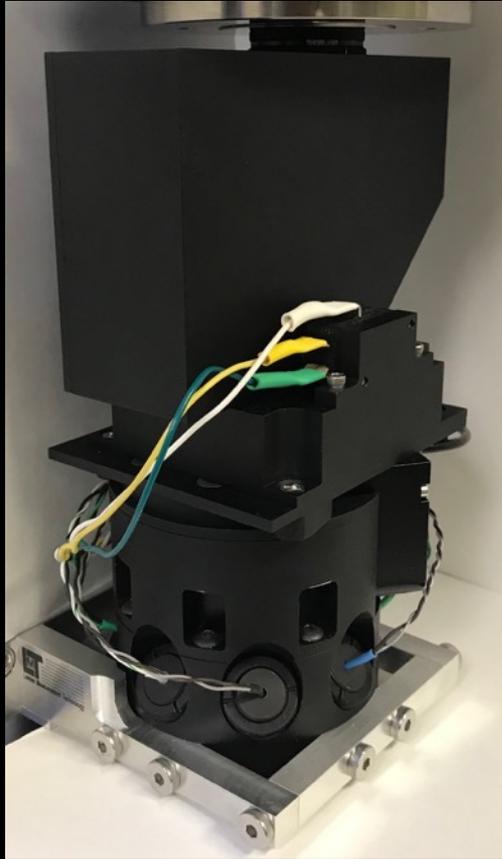


Z-Stacked Image

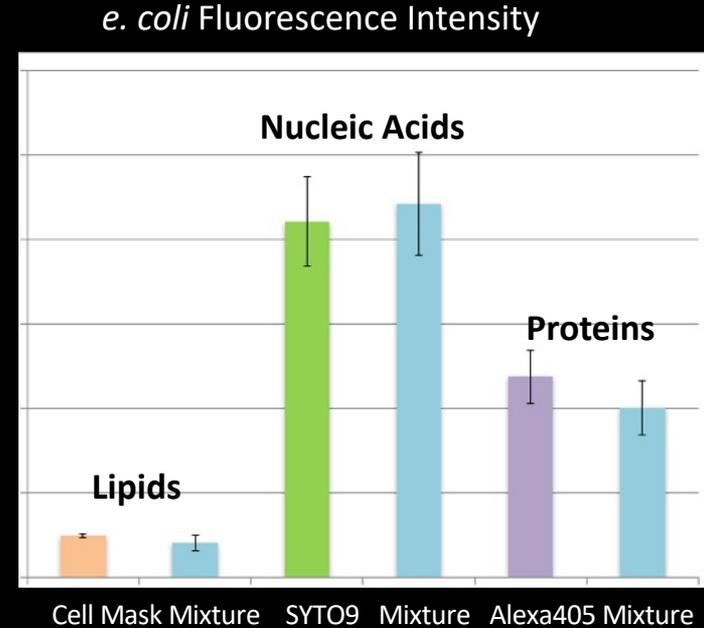
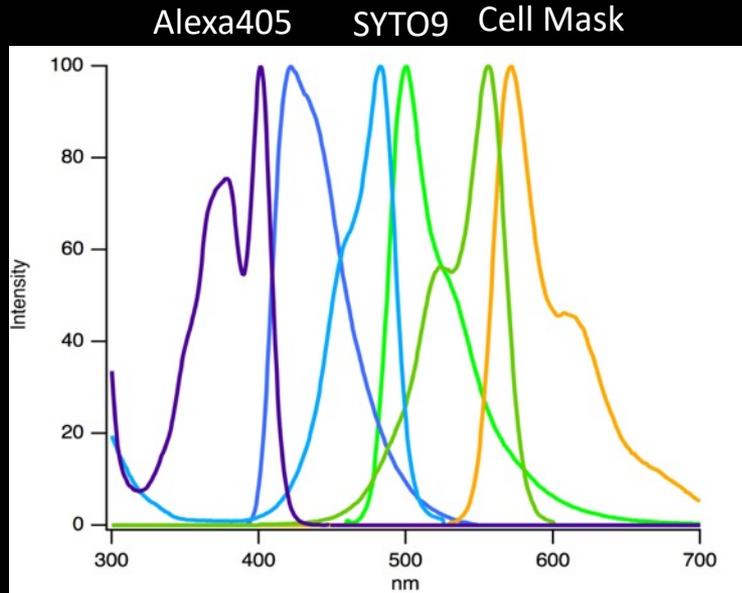
- *Bacillus megaterium*
- SYBR Gold
- Nucleic Acid Stain
- width $\sim 1.5 \mu\text{m}$



Automated Image Z-Stacking



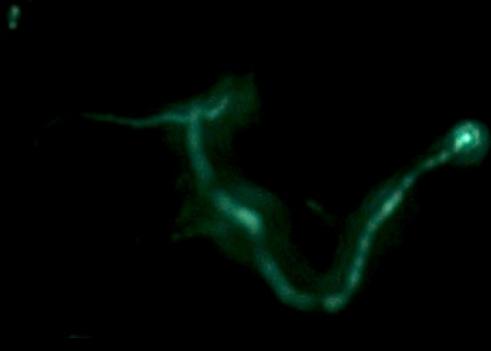
Fluorescent Probes



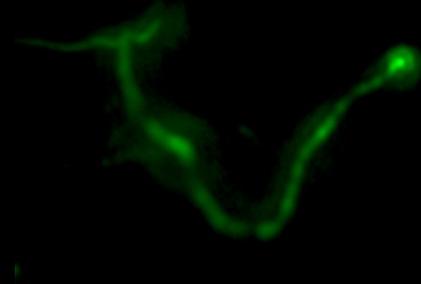
- 3-Stain Combination – Targets – Proteins, Lipids, Nucleic Acids
- No loss of fluorescent intensity for 3-dye mixture vs. individual dyes
- All targets stained simultaneously after capture on SiN filters
- Detection of stained structures below spatial resolution limit

Fluorescent Probes

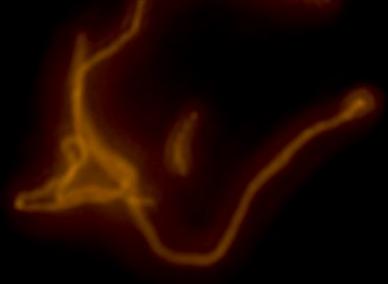
P. boydii ~ 1 Micron Wide



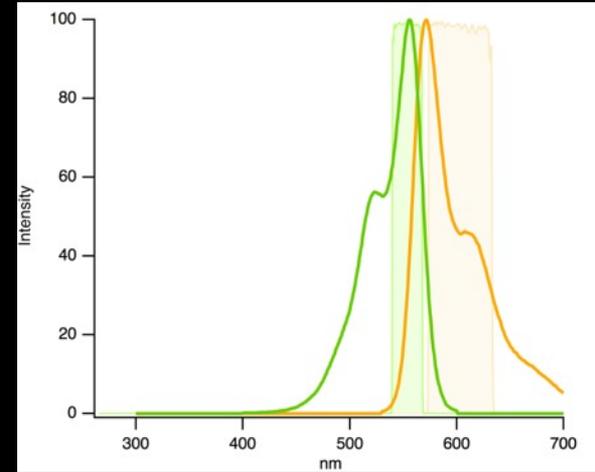
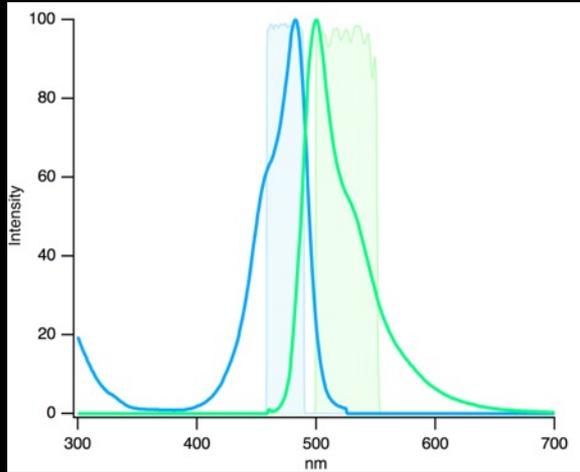
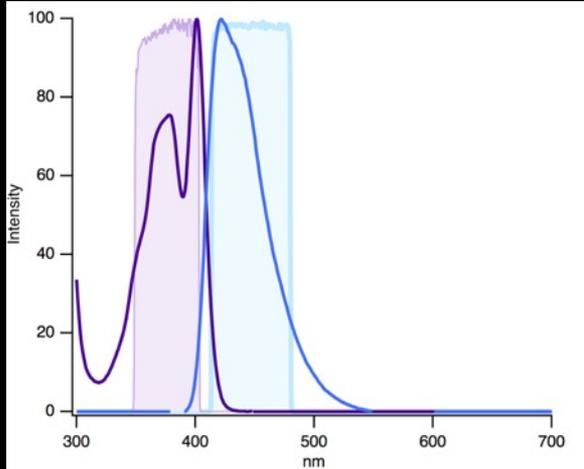
Alexa Fluor 405 - Membrane Proteins



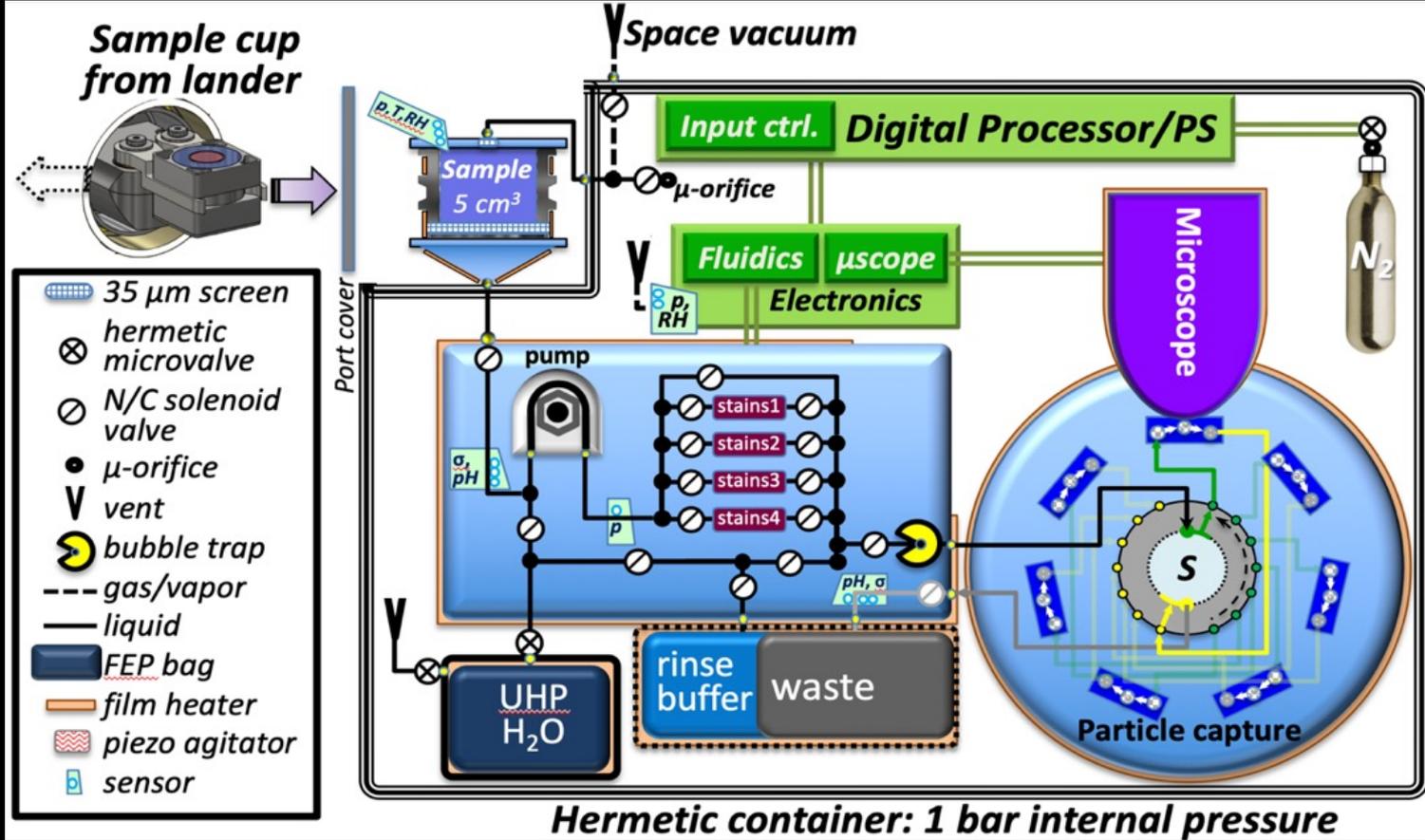
SYTO9 - Nucleic Acids



Cell Mask - Membrane Lipids



ELM Block Diagram

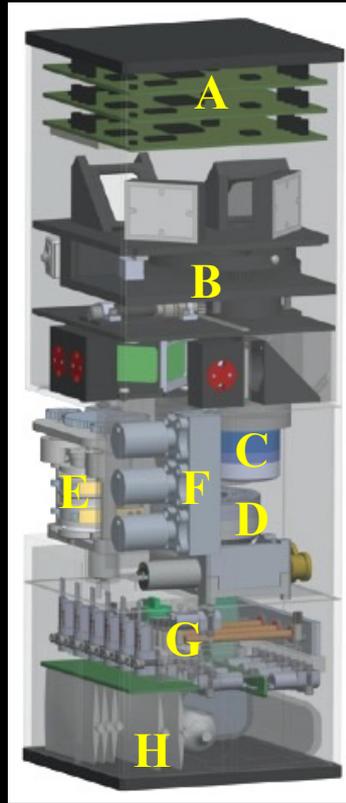


ELM Subsystems and ICEE-2 Development

Lineage: FLAIR Prototype (CubeSat) & LiFE Prototype (COLDTech)

Subsystems:

- A) Camera Board/Electronics
- B) Optics (Microscope)
- C) Microscope Objective
- D) Sample Filter Stage
- E) Sample Transfer Dock
- F) Hermetic Valve Block
- G) Fluidics Manifold
- H) Fluid Storage/Waste



ICEE-2 Focus:

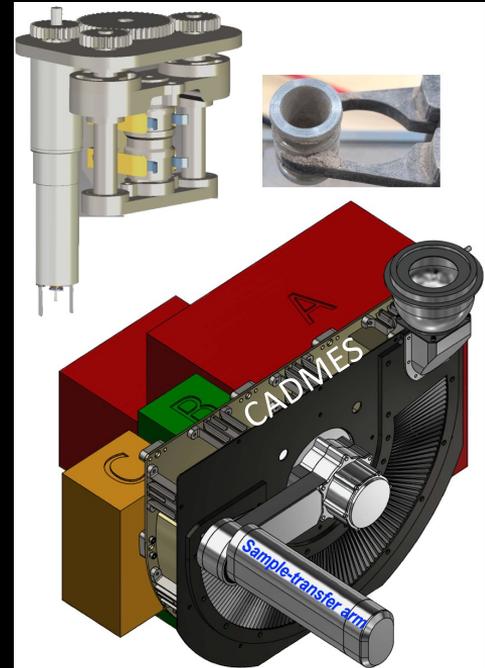
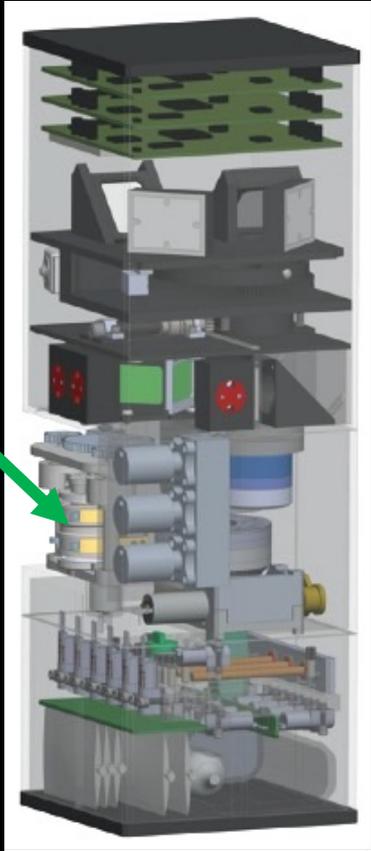
- Environmental
- Packaging
- Performance
- COTS → Custom

- 1) Sample Transfer
- 2) Sample Filter Stage
- 3) Objective and Piezo Stage
- 4) Rad-Tolerant Camera
- 5) ConOps & Data Processing

Sample Transfer Subsystem

Key Features

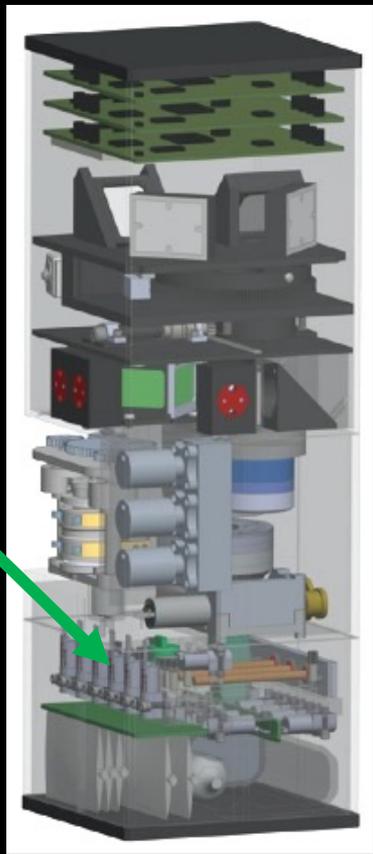
- Designed for compatibility with the CADMES (Collaborative Acceptance and Distribution for Measuring European Samples) system (C. Malespin, PI GSFC)
- 5 cc sample cup w/mesh bottom; sealing mechanism for reversible docking and undocking; thermal control to melt ice sample; piezo-agitator for particle transfer, coupled with fluidic manifold transfer line
- Honeybee Robotics breadboard complete.
- Brassboard including environmental testing in progress.



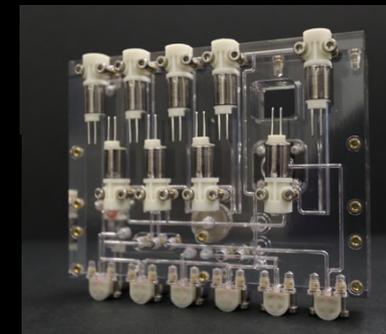
Fluidics Subsystem (Sample Processor)

Key Features

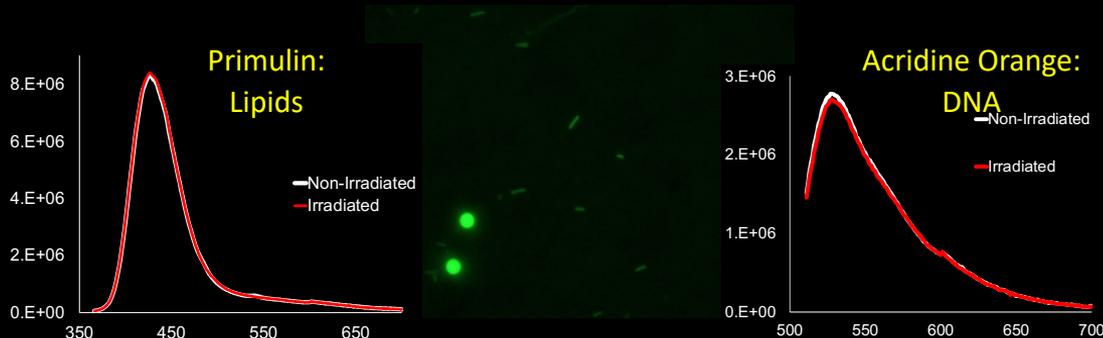
- 8 cm x 9 cm Monolithic Fluidic Manifold
- Space Biology Lineage:
Requirement for Perfect Sterility
- pH, ORP and Pressure Measurements with feedback control
- Fluorescent Stains: Porous Polymer Stabilization and Storage
- Radiation Tested to > 300 krad; including fluorescent stains



"Top"

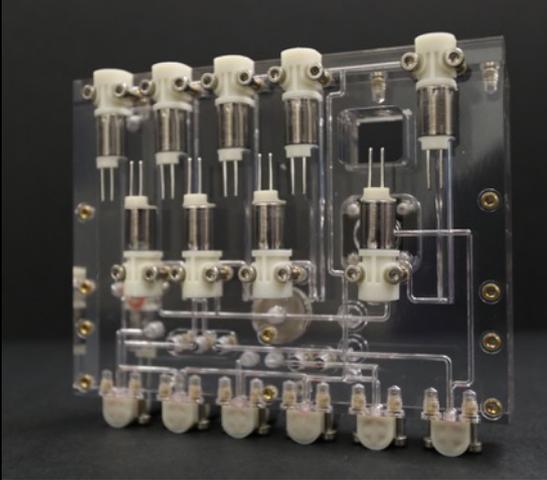


"Bottom"

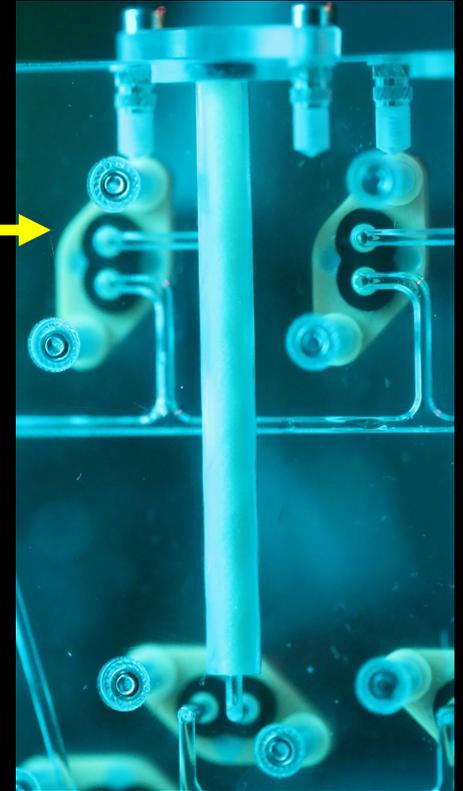
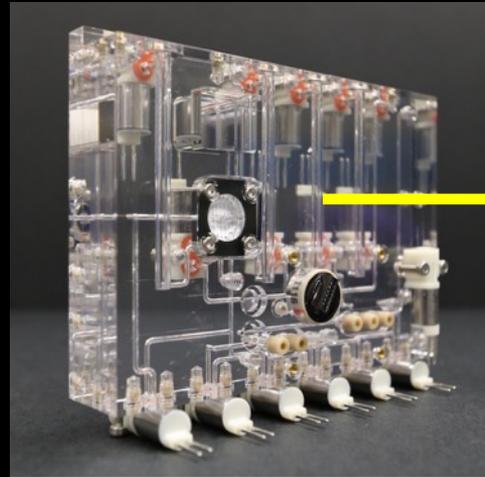


Fluorescent Probe Storage and Mixing

"Back"



"Front"



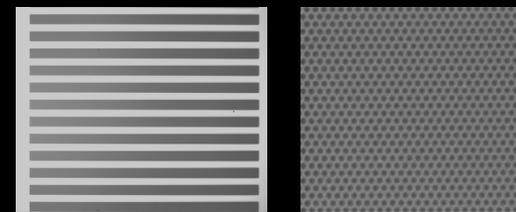
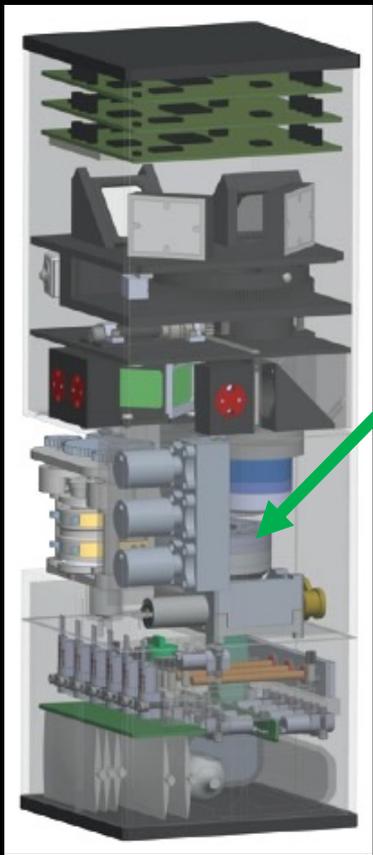
Key Features

- 8 cm x 10 cm Monolithic Fluidic Manifold
- Space Biology Lineage: Requirement for Perfect Sterility
- Radiation Tested to > 300 krad
- pH, ORP and Pressure Measurements with feedback control
- Fluorescent Stains: Porous Polymer Stabilization and Storage

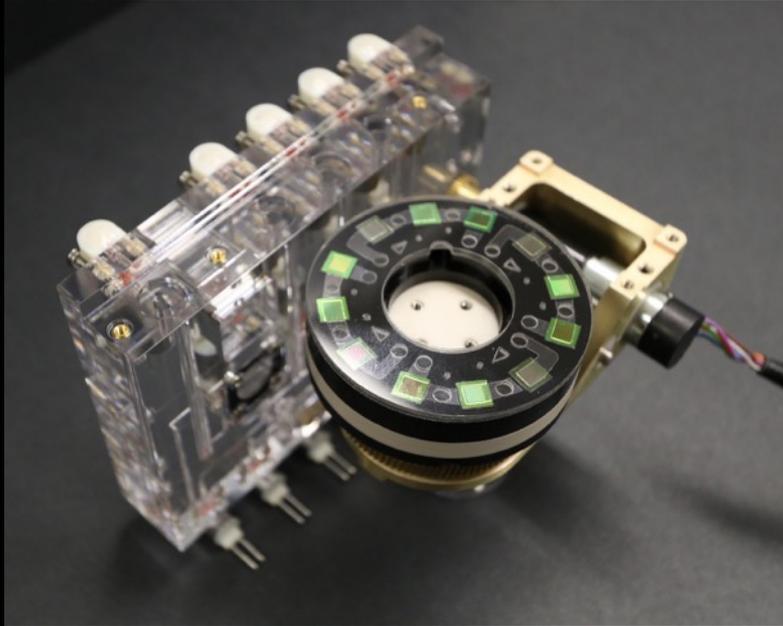
Fluidics Subsystem (Particle Filter Stage)

Key Features

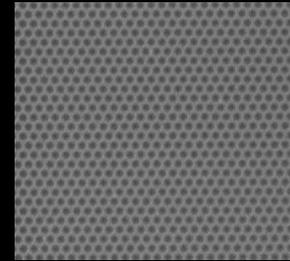
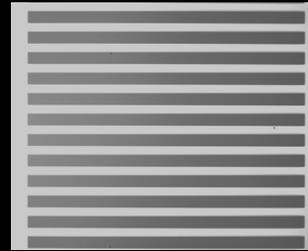
- Custom silicon nitride membrane particle filters on Si wafer
- 7-sets of three inline filters
- Three stage filtering 10 μm ; 1 μm ; 0.15 μm
- Etched calibration targets on Si wafer
- Filter geometry optimized for microscope field of view
- Optical positioning sensor on rotary stage



Silicon Nitride Membrane Particle Filtering and Directional Concentration



- Three stage filtering 10 μm ; 1 μm ; 0.2 μm
- 5 mm x 5 mm Si Frame
- 3 mm x 3 mm SiN Filter Collection Area
- 14 X 50 μm x 3 mm collection fields
- Allows for directional particle concentration
- Provides optimized imaging efficiency

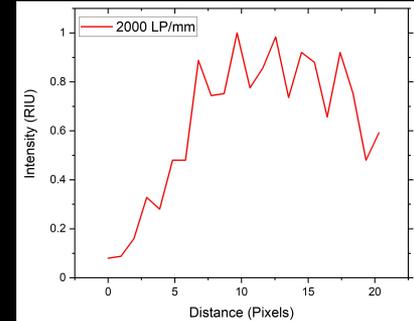


Time

Optics Subsystem (Microscope)

Key Features:

- Custom 40x objective
- Field-of-View - 300x263 μm
- Spatial Resolution < 0.5 μm
- Depth-of-Field - 2 μm
- Custom piezo focusing stage
- Z-motion up to 400 μm
- Z-positioning accuracy 1 nm
- LED Excitation Wavelengths (4):
275, 375, 470, & 525 nm
- Emission Filters (4):
334, 470, 529, & 579

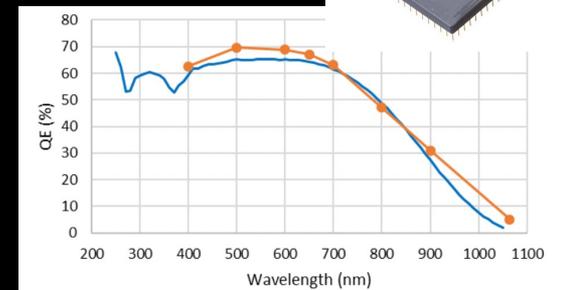
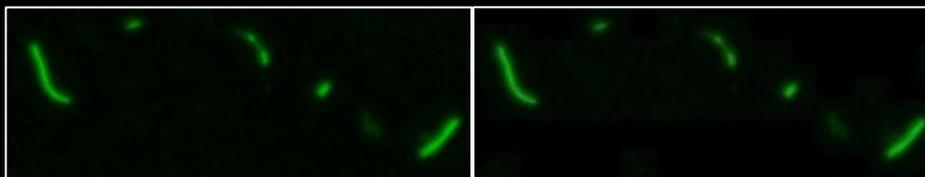
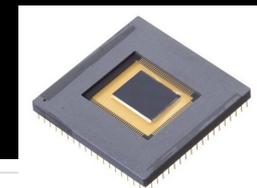
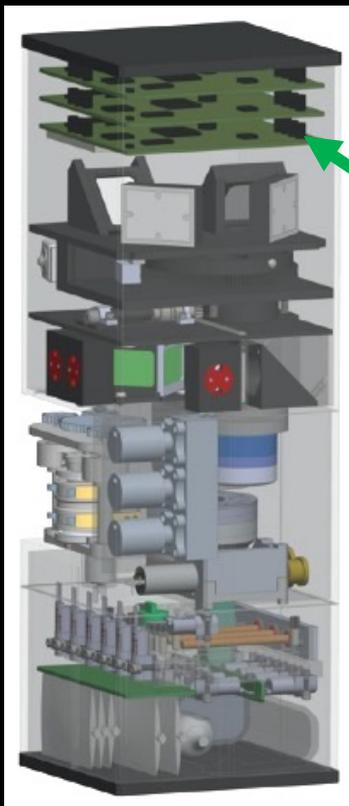


Optical components have passed radiation testing to 300 krad, including:
Bandpass filters (single- and multi-band); LED modules; Lenses and Lens Materials.

Electronics Subsystem (& Microscope Camera)

Key Features:

- COTS Camera replaced with custom board using e2V CIS115 image sensor (JUICE mission heritage)
- Back thinned - UV sensitive
- Sample maps to $0.18 \mu\text{m}/\text{pixel}$
- Image processing software ported to rad-hard microcontroller
- **Autonomous Image Processing:** incorporates z-stacking, data interpretation, data compression and decisional data generation



Acknowledgements

NASA Instrument Concepts for Europa Exploration