PRESENTATION ABSTRACT

Biofilms On Orbit and On Earth: Current Methods, Future Needs

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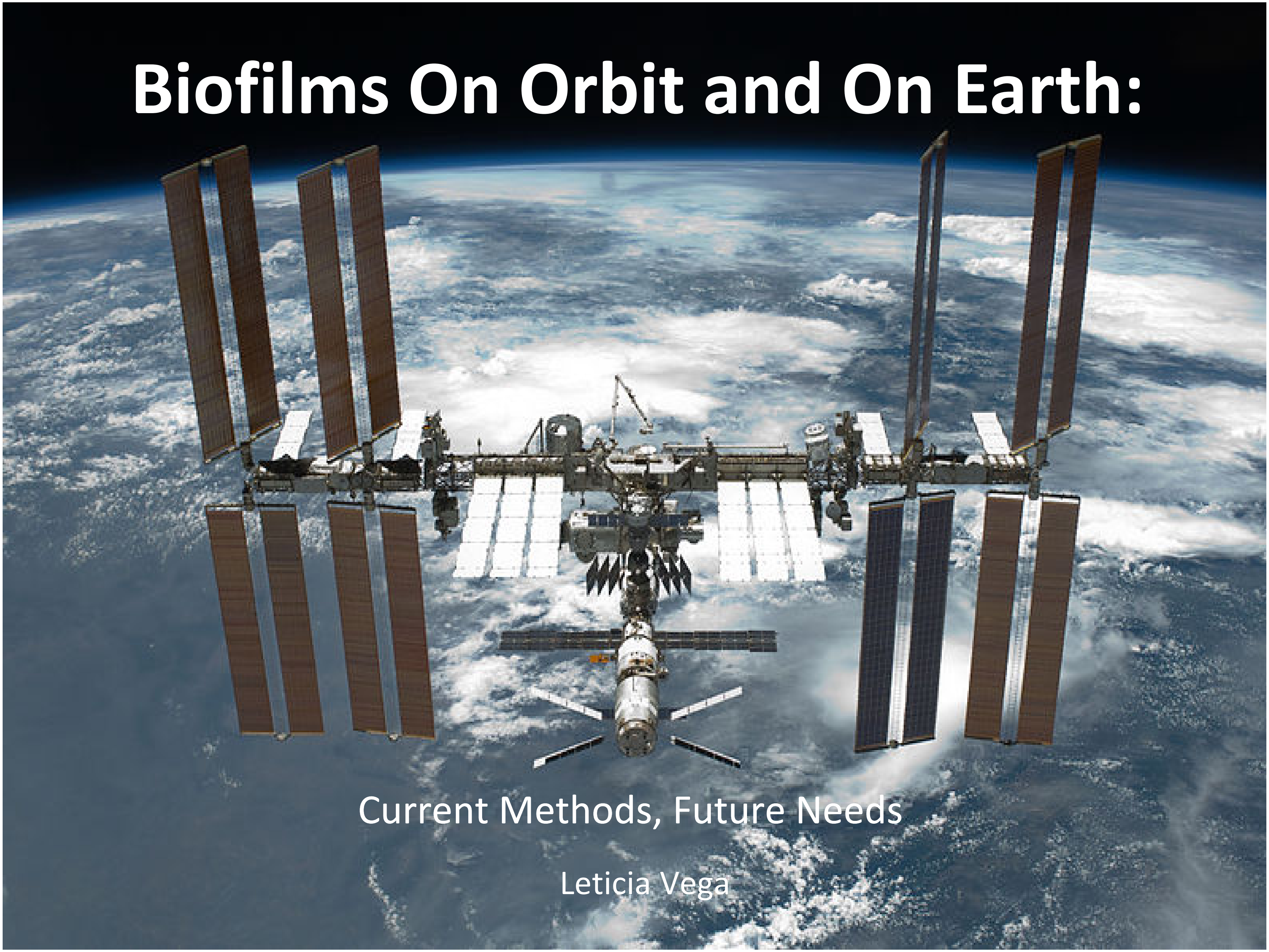
Biofilms have played a significant role on the effectiveness of life support hardware on the Space Shuttle and International Space Station (ISS). This presentation will discuss how biofilms impact flight hardware, how on orbit biofilms are analyzed from an engineering and research perspective, and future needs to analyze and utilize biofilms for long duration, deep space missions.

Biofilms On Orbit and On Earth:

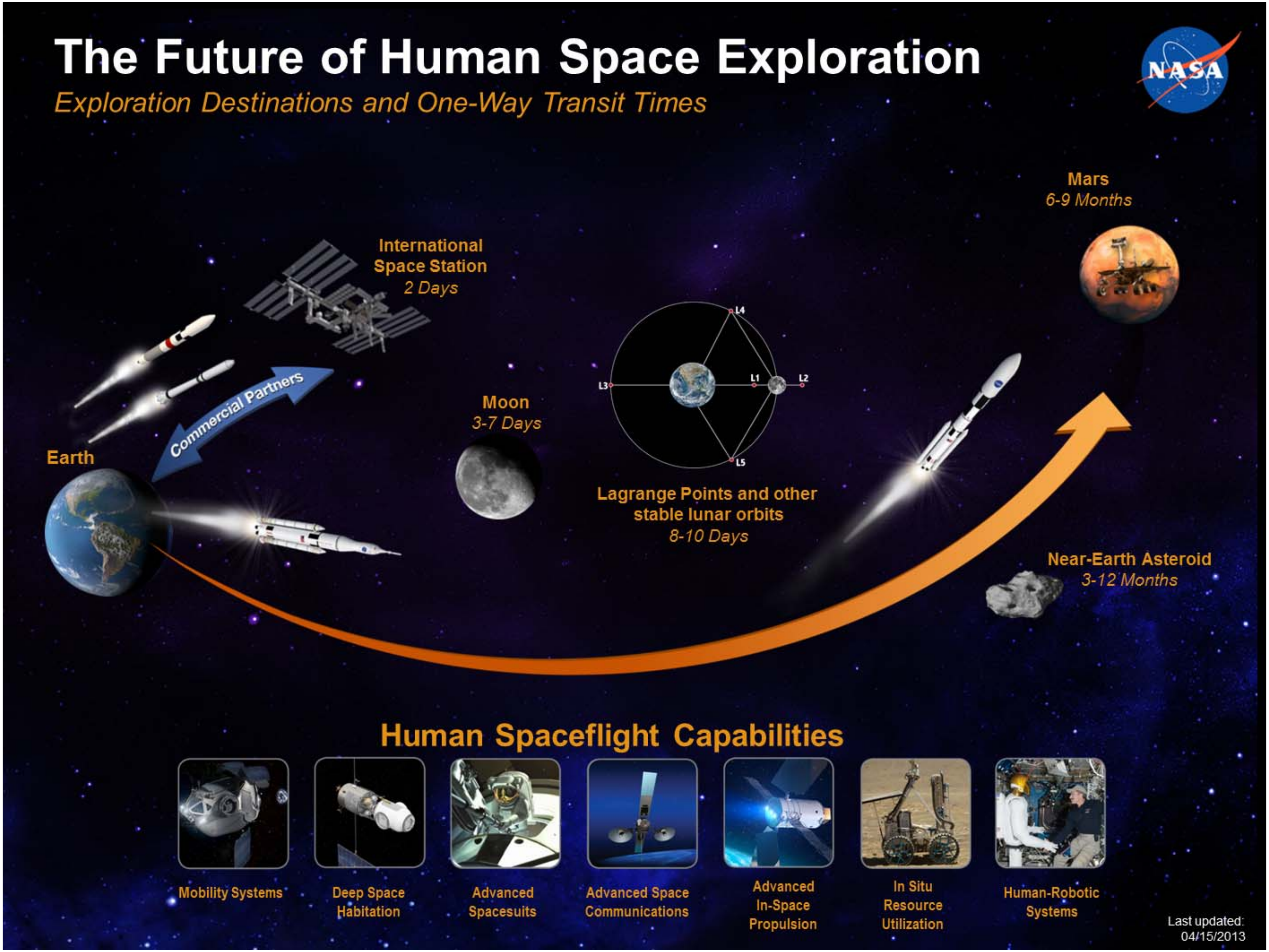


Current Methods, Future Needs

Leticia Vega

Overview

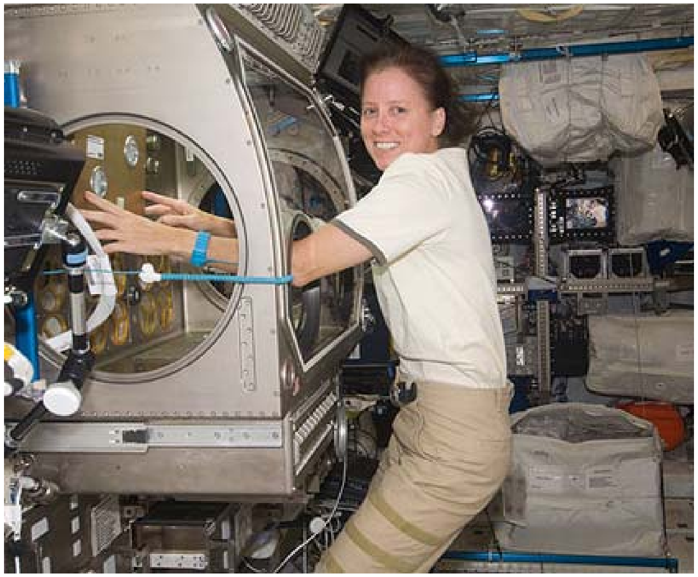
- Impact of biofilms on space exploration
- Biofilm research
  - On orbit
    - Space Shuttle
    - ISS
  - On earth
- Impact of biofilms flight hardware
- Lessons learned
- Next steps/ future needs



The Future of Human Space Exploration
Exploration Destinations and One-Way Transit Times
NASA
International Space Station
2 Days
Commercial Partners
Earth
Moon
3-7 Days
L4
L3
L1
L2
L5
Lagrange Points and other stable lunar orbits
8-10 Days
Mars
6-9 Months
Near-Earth Asteroid
3-12 Months
Human Spaceflight Capabilities
Mobility Systems
Deep Space Habitation
Advanced Spacesuits
Advanced Space Communications
Advanced In-Space Propulsion
In Situ Resource Utilization
Human-Robotic Systems
Last updated: 04/15/2013

Challenges Associated With Analysis of Biofilms in Space

- Closed system
  - Working volume of ISS: 29,600 ft^3
    - Size of a house
- Small fluid volumes
  - ~2m^3 water volume on ISS
- Time constraints
  - Daily activities
  - Station maintenance
  - Research

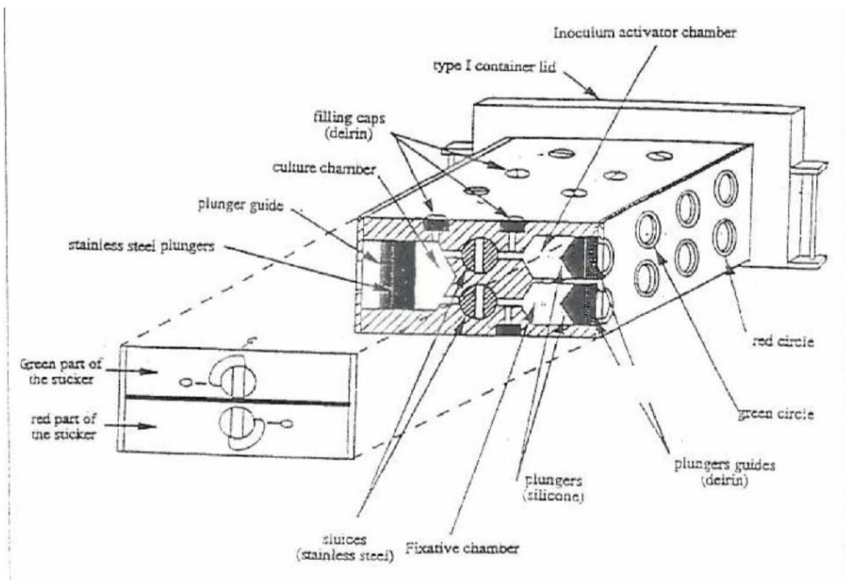


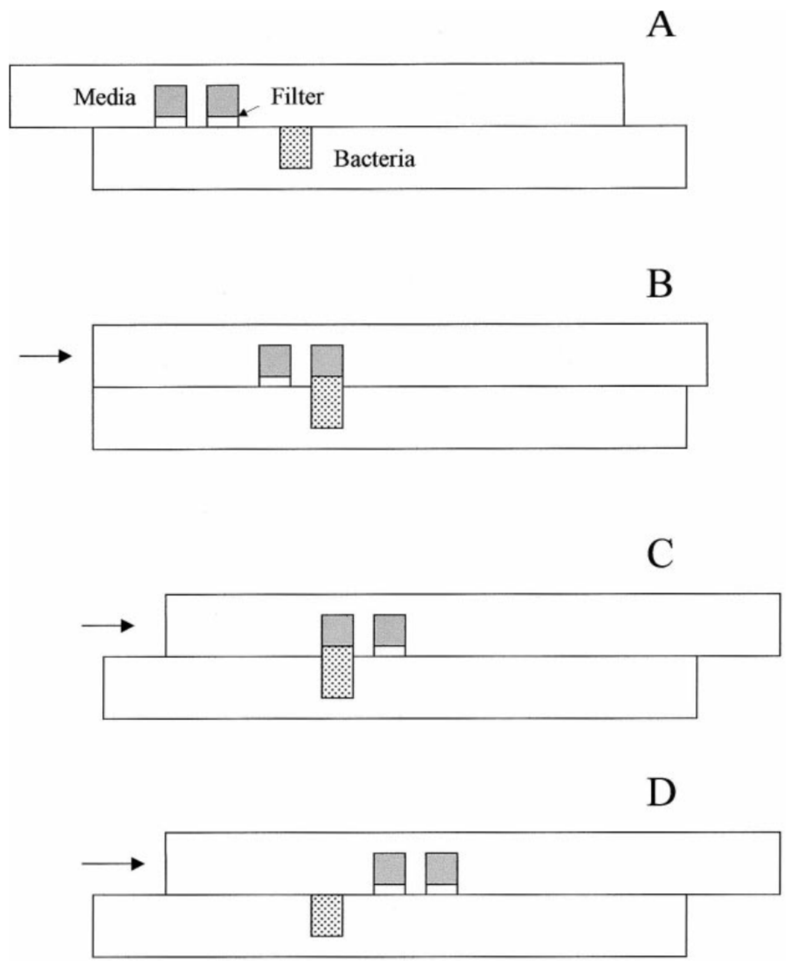
Biofilm Research

Characteristics of a Bacterial Culture in Microgravity

- Typical physical stresses are absent
  - Low shear forces
  - No convection forces
- Shorter lag and longer log phase growth
- Increased resistance to antibiotics

Examples of Devices Used in Spaceflight





Spaceflight Biofilm Experiments

- Pyle (STS-81, 1997)
  - Compared on-orbit biofilm formation and growth *Burkholderia cepacia* to a parallel experiment performed at KSC
- McLean (STS-95, 1998)
  - Evaluated the effect of microgravity on *Pseudomonas aeruginosa* biofilms
- Wilson et al. (STS-115, 2006)
  - Compared colony morphology and expression of virulence genes in *Salmonella typhimurium* to a parallel experiment
- Kim et al. (STS-132 and-135, 2010 and 2011)
  - Evaluated the growth *Pseudomonas aeruginosa* biofilms on orbit and determined that there is an altered biofilm morphology as compared to those growth in identical conditions on orbit.

Mimicking Microgravity: Rotating Wall Vessels

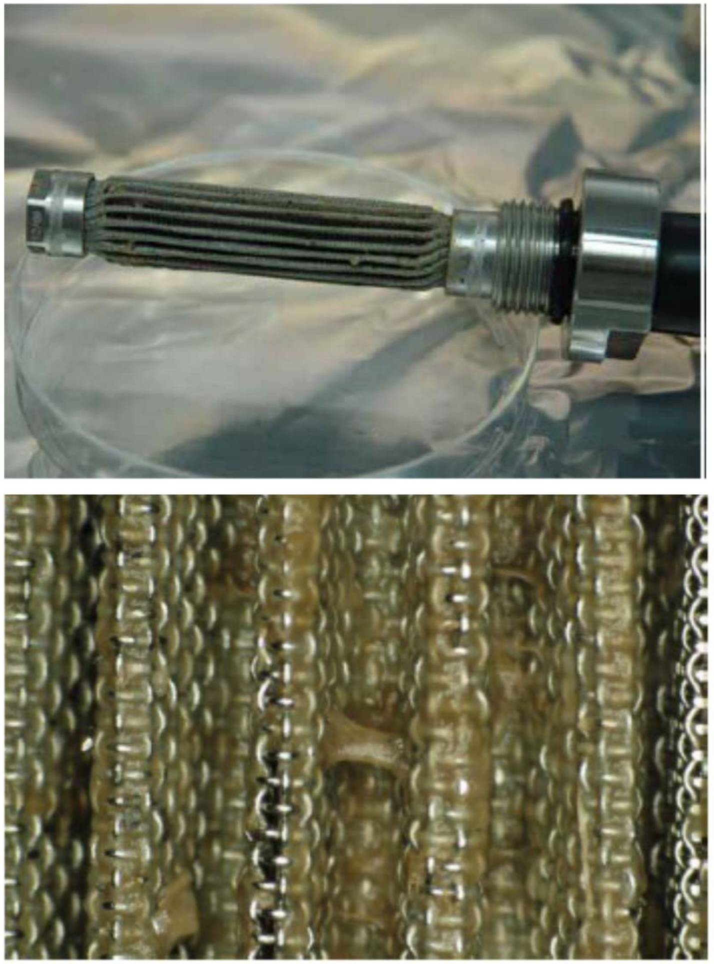
- Developed at JSC to perform mammalian cell culture experiments
  - Axis of rotation is perpendicular to the gravity force, mimicking microgravity
- Modified for biofilm research
  - Lynch, et al. 2006 (*E.coli*)
  - Crabbe, et al 2010 (PAO-1)

Source: http://www.synthecon.com/autoclavable-vessel-batch-systems.html

Impacts of Biofilms to Flight Hardware

Biofilms and Flight Hardware

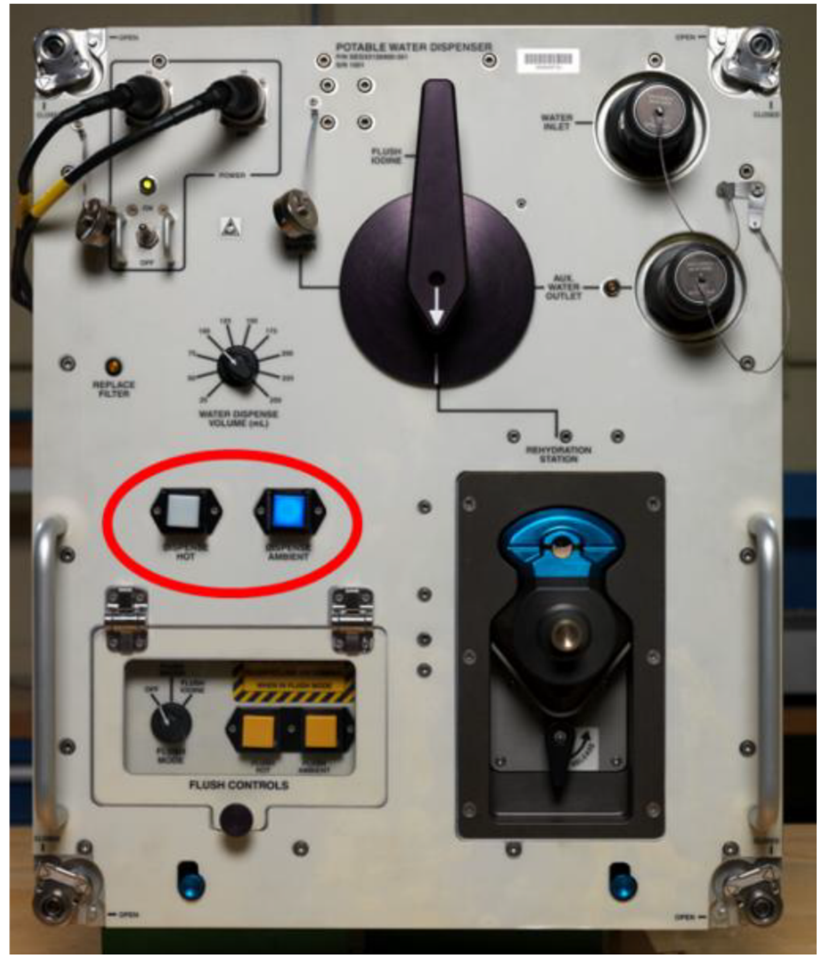
- Biofilms impact flight hardware by
  - Affecting fluid quality
  - Fouling fluid lines
  - Decreasing lifetime of hardware and hardware components



From “Microbial Characterization of the International Space Station Water Processor Assembly External Filter Assembly ” AIAA 2012-3595, 2012

Example #1: A Potable Water Biofilm

- Potable Water Dispenser (PWD)
  - Provides drinking water to the crew for consumption and food rehydration
  - Source water is recycled water from humidity condensate and urine
    - Bacterial limit: 50 CFU/mL



From “International Space Station United States On-orbit Segment Potable Water Dispenser On-orbit Functionality vs. Design” AIAA 2010-6250, 2010

- Issue
  - Counts exceed NASA potable water limits
- Cause
  - System was wetted with non sterile water prior to launch and left stagnant for 6 months
  - Exacerbated by corrugated tubing in PWD
- Flight Solution
  - Flush with iodinated water

From "International Space Station United States On-orbit Segment Potable Water Dispenser On-orbit Functionality vs. Design" AIAA 2010-6250, 2010

The Making of a Space Biofilm

- Scientists at JSC inoculated several stainless steel hoses with a consortium of bacteria
  - Mimic the type of biofilm likely present in the PWD
- Challenged biofilm with several disinfectants to identify potential alternative biocides
  - 6% hydrogen peroxide, colloidal silver were effective

Example #2: Serendipitous Finding of a Biofouling Resistant Heat Exchanger

- Service and Performance Check Out Unit Heat Exchanger (SPCU HX)
  - Provides thermal control to the spacesuits before and after a spacewalk
  - Issues
    - Changes in coolant chemistry
    - Increased bacterial concentration

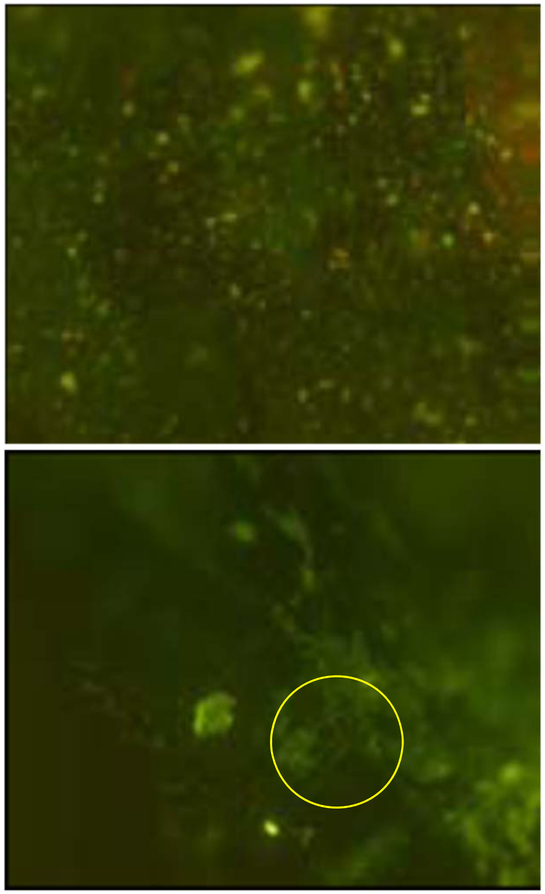
From "Inhibition of Biofilm Formation on the Service and Performance Heat Exchanger by Quorum Sensing Inhibition" SAE 2007-01-3143, 2007.

Analysis of Heat Exchanger

- Heat exchanger was replaced and analyzed for biofilm & MIC
  - Evaluated using
    - Scanning electron microscopy (SEM)
    - Epifluorescence microscopy
    - Auger microscopy
- Results
  - Planktonic growth <u>without</u> biofilm

Verification of Biofilm Inhibition

- Objective: to confirm biofilm inhibition on heat exchanger materials
- Method
  - Coupons were inoculated and incubated for 10 weeks
  - Samples were collected for
    - Heterotrophic plate count
    - Microscopy analysis
- Results
  - No microcolony formation



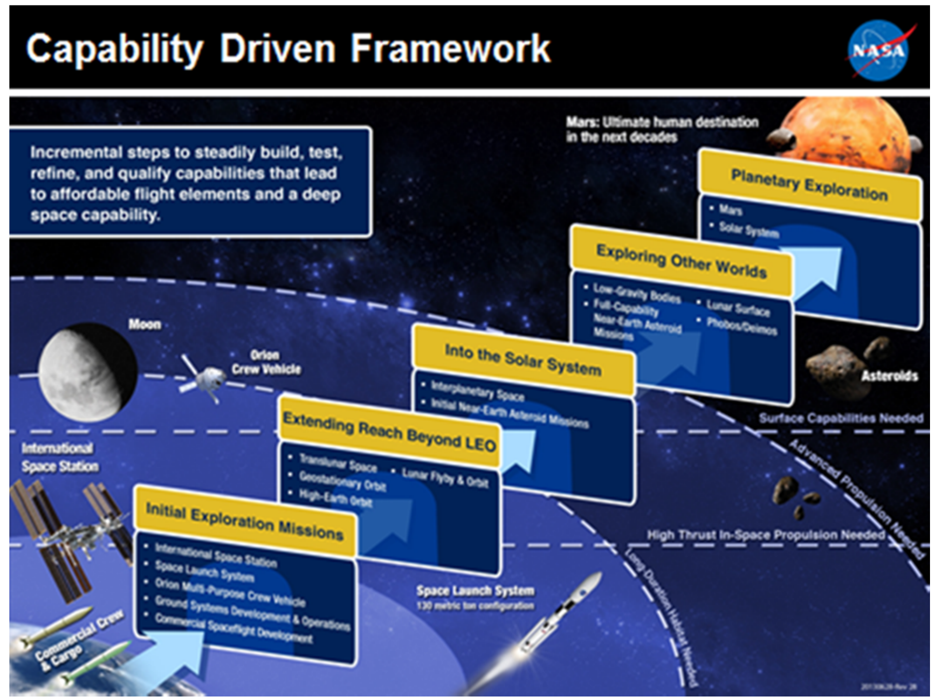
Microscopy of control (top) and SPCU HX (bottom) coupons test day 65.

Lessons Learned

- Research
  - Biofilm growth is prolific in spaceflight and can subsequently impact virulence
  - Microgravity can affect biofilm morphology
- Practical
  - “Start clean, stay clean”

Future Needs for Long Duration Spaceflight

- Water quality verification
  - Prevent problems before they start
- Ability to recover from an event
  - Non-toxic
  - Fast acting
  - Regenerative



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