Cell Science-03 (CS-03) Payload Overview

POIWG #43

April 26, 2018

David R. Segovia

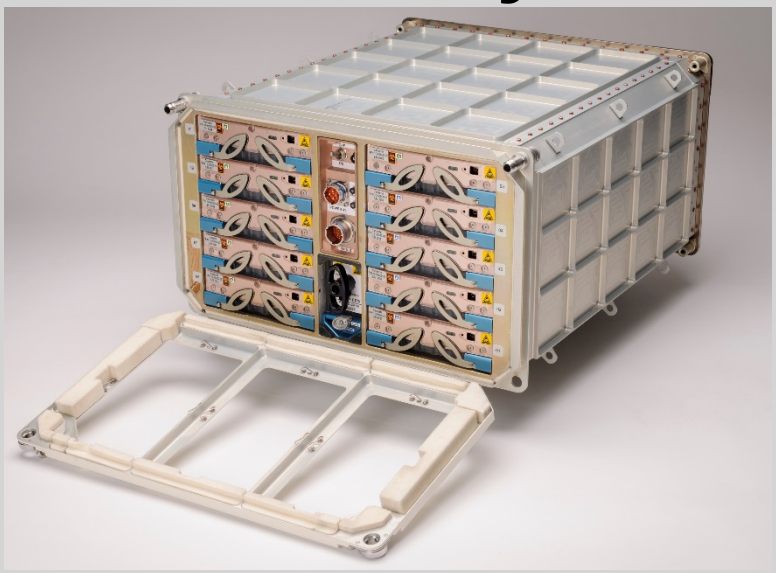
Ames Research Center (ARC)

Thank you!

NASA SILICON VALLEY

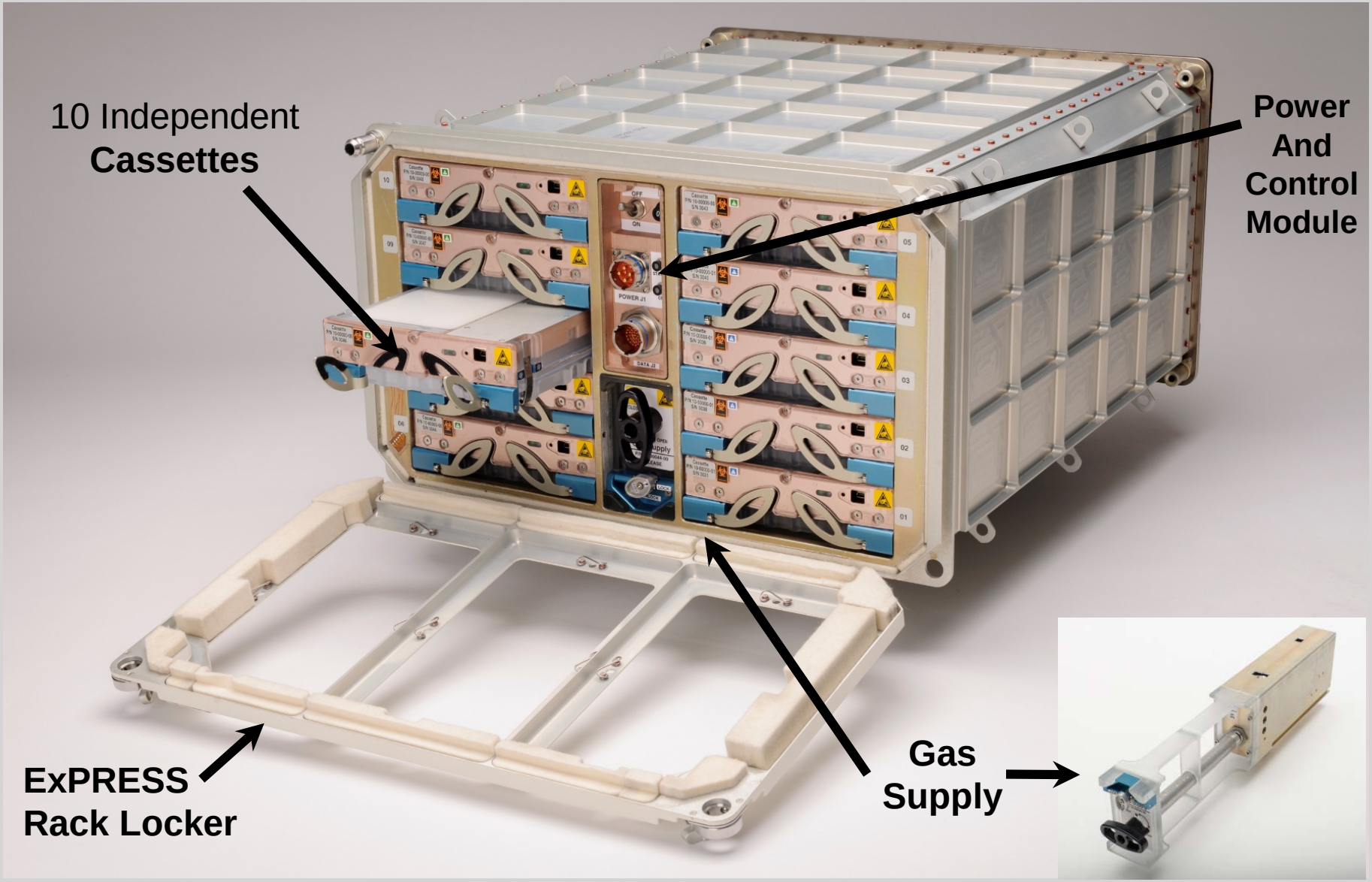
AMES RESEARCH CENTER

Bioculture System



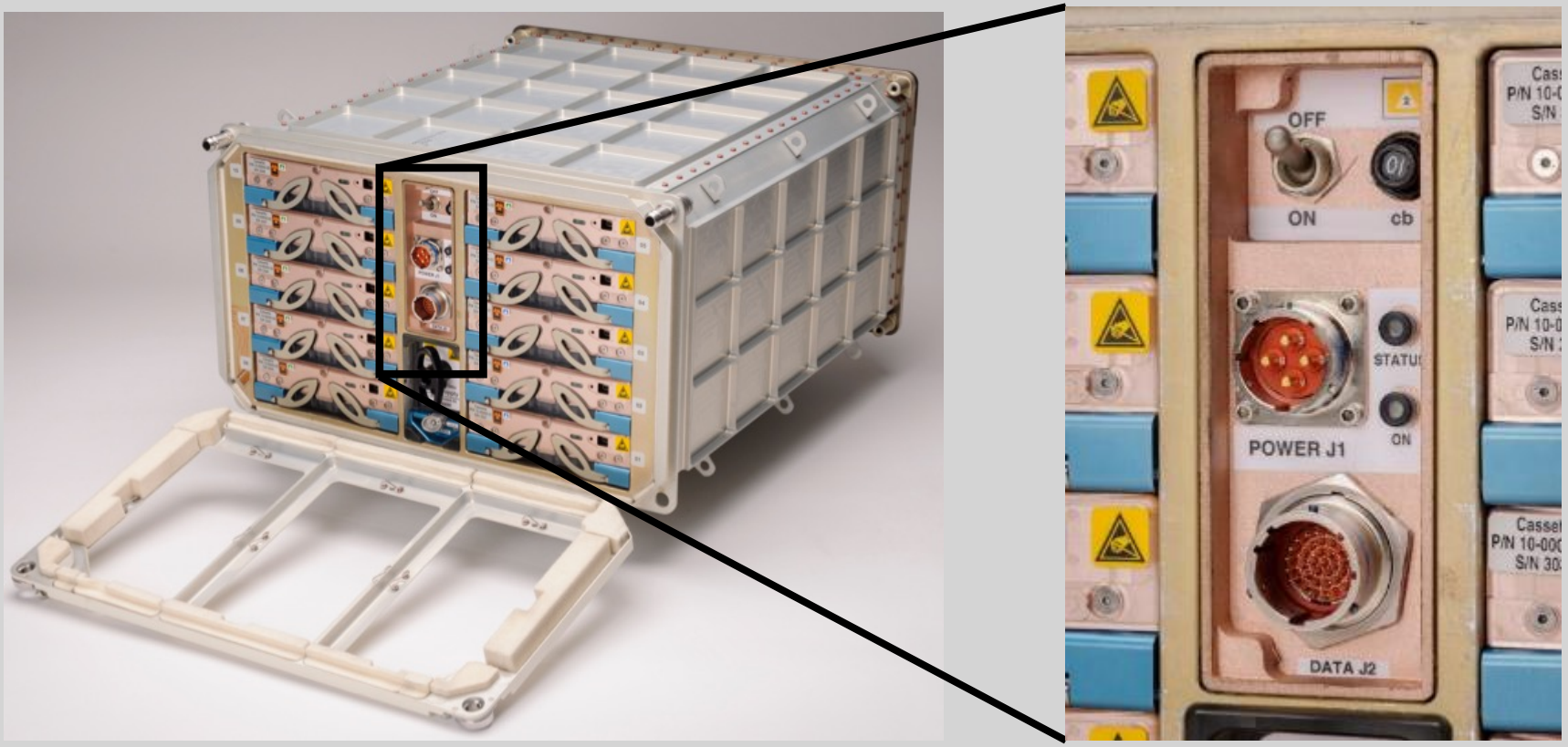
- Automated cell biology system with 10 independent cassettes
- Programmable manipulations include:
  - Media flow rate and feeding schedule
  - Adjustable gas dosing
  - Sample collection
  - Fixative injection
- Cassettes will be removed and opened in Life Sciences Glovebox (LSG) to allow initiation of cell cultures, media changeout, sample retrieval and preservation.

Bioculture System

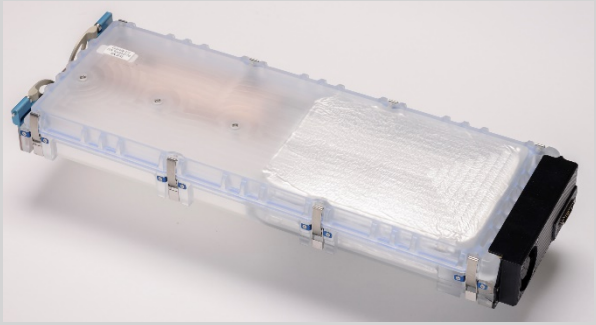


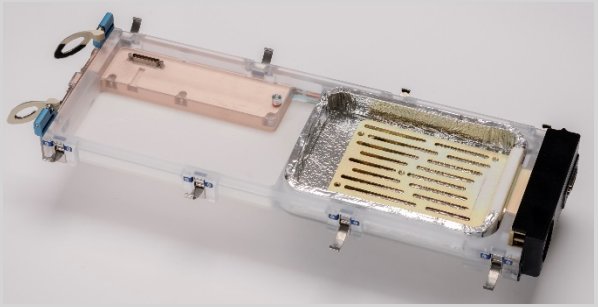
Power and Control Module

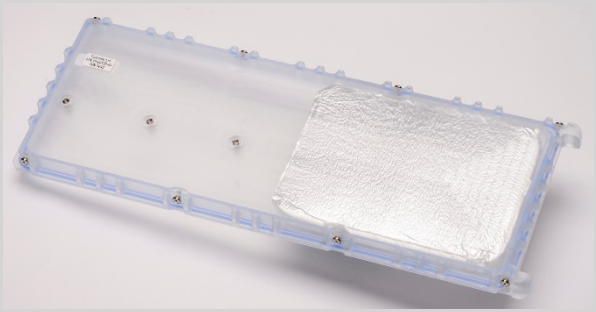
The Power and Control Module takes 28V Rack voltage, converts it to 12V, and uses standard interfaces for both power and data to the Rack. The Power and Control Module contains an On/Off toggle switch, a circuit breaker, Data Connector, Power Connector, and two LEDs for Power/Fault Status.

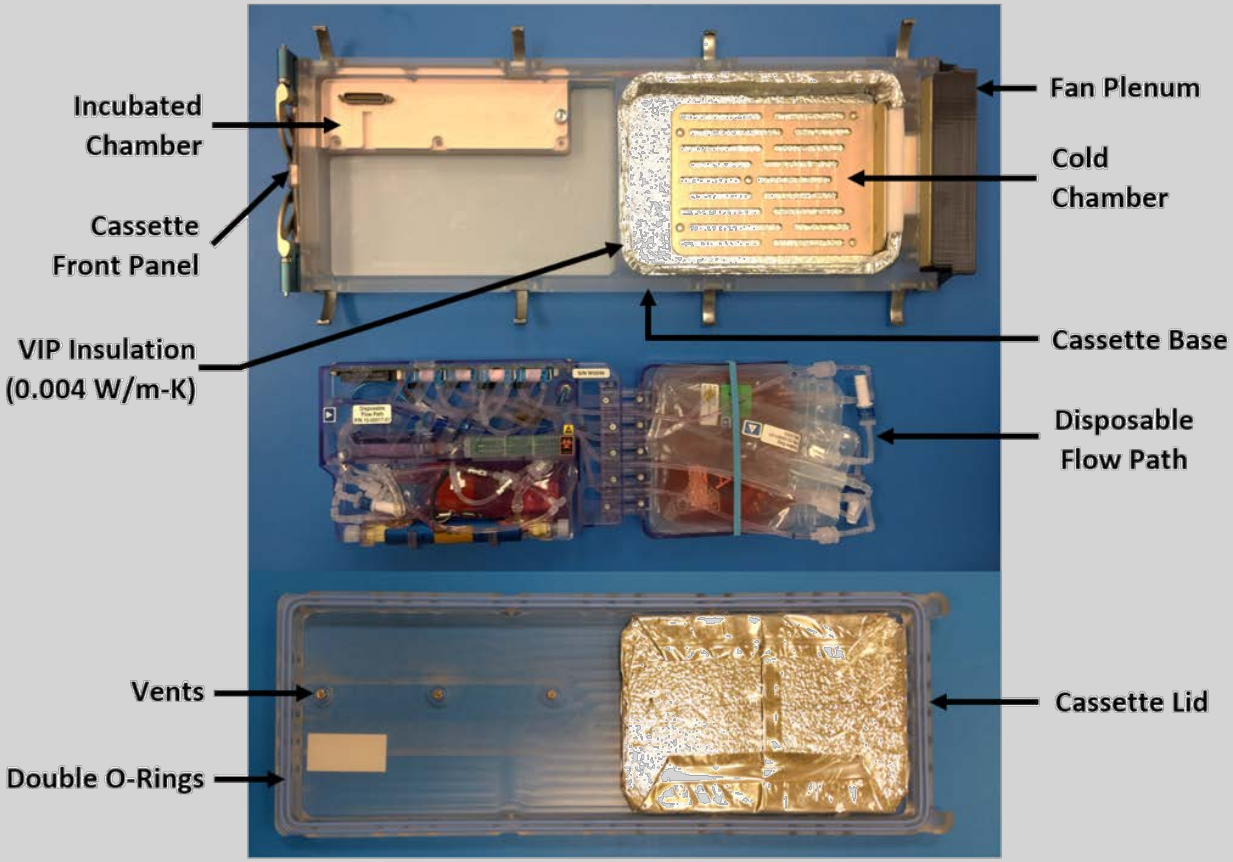


Durable Cassette with Disposable Flow Path

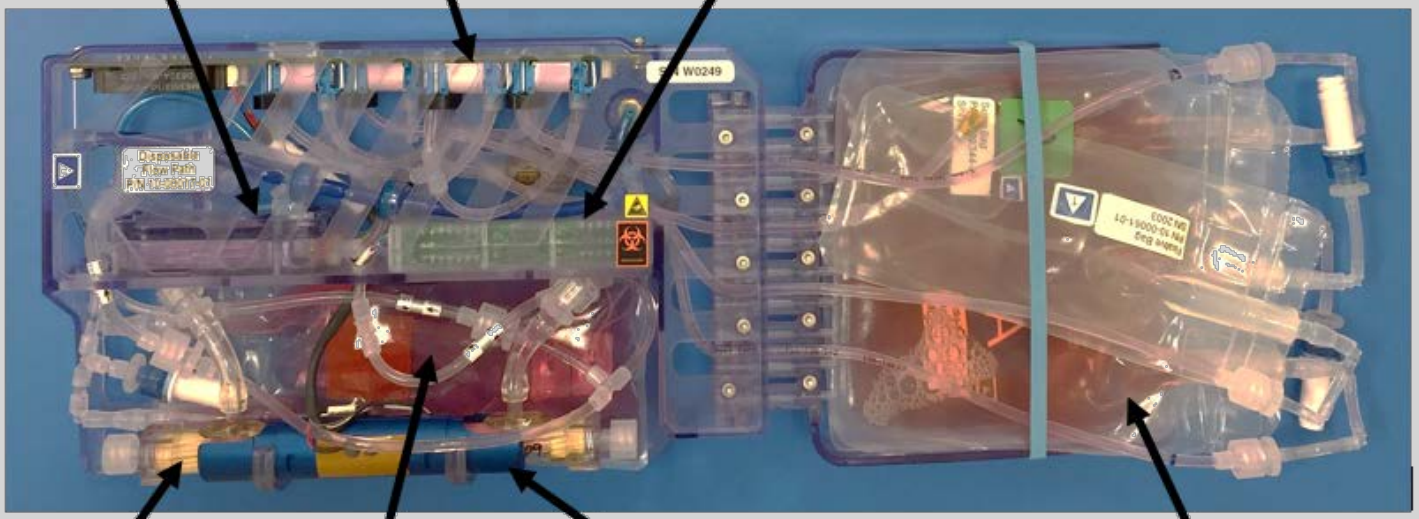








Disposable Flow Path



CS-03 Experiment Summary

Principal Investigators	Elizabeth Blaber, Ph.D. Universities Space Research Association/ NASA Ames Research Center
Co-Investigators	Eduardo Almeida, Ph.D NASA Ames Research Center
Sponsor	SLPSRA – Space Biology Program
Funding Authority	NASA / Human Exploration & Operations / ISS NASA / Human Exploration & Operations / Space Life and Physical Sciences
Experiment Title	Osteogenic Differentiation of Somatic Stem Cells in Space: A Study Investigating the Role of CDKN1a/p21 on Mesenchymal Stem Cell Proliferation, Differentiation, and Regeneration in Microgravity.
Experiment Duration	30 Days – Initiated on orbit
Cell Type	Primary mouse bone marrow mesenchymal cells
Independent Variable	Whether cells are from Wildtype (WT) or CDKN1a/p21 knockout (KO) mice

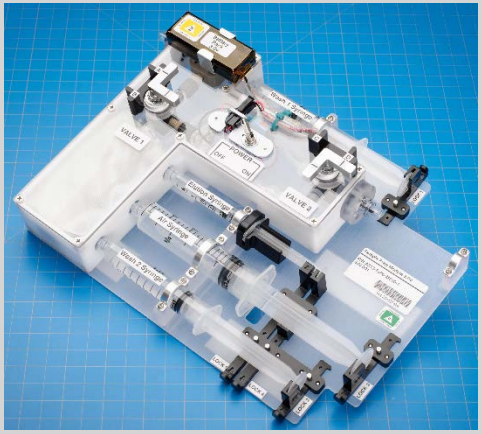
New Crew Operations

Disposable Flow Paths:

- Disposable Flow Paths will be flown separately and the crew will install into the BCS Cassettes on-orbit.

WetLab RNA SmartCycler:

- System enables on-board space biology gene expression research as well as analysis of environmental containments
- System will be used at two time points to sample cells to verify culture growth genes and final fixation.



Bioreactor Injections:

- Frozen cells derived from two types of mice will be injected into the Bioreactors.

MinION:

- Portable real-time device for DNA and RNA sequencing.

CS-03 Timeline

Turnover Unpowered Bioculture System (BCS) — L-9w

Ship soft stow items to JSC — L-6w

Turnover cold stowage items (media bags, factors, and frozen cells) for use on Orbit — L-2d

SpX-16 Launch

Set up LSG — NLT L+5d

Dragon Berth

Thaw Frozen Cell Cultures and Inject into Bioreactors

Will refer to this as "Initiation" day

Bag Change Out — TBD

Gas Supply Change Out #1 — TBD

Sample Cells from Bioreactors for WetLab2 analysis #1 — Initiation+10d

Bag Change Out — TBD

Gas Supply Change Out #2 — TBD

Bag Change Out — TBD

Gas Supply Change Out #3 — TBD

Bag Change Out — TBD

Sample Cells from Bioreactors for WetLab2 analysis #2 — Initiation+20d

Automated Fixative Injection — Initiation+30d

Remove Bioreactors and all bags from Cassettes and put in Cold Stowage. — Initiation+31d

Tear Down LSG — Flexible

Transfer Bioculture System, cold and soft stow items to Dragon — U-2d

SpX-16 Splashdown

Retrieval of cold stow items — TBD

Priority Return of Soft-stowed BCS to ARC — 2-5 weeks

Purple Text indicates the activity occurs in the LSG

Integration Overview

- **Bioculture System will be operational for first 30 days after SpX-16 launch**
- **Bioculture System Facility Payload Integration Agreement (PIA) Letter contains the following unique agreements:**
  - Bioculture System shall remain on the pad no longer than 48hrs after turnover, after which the biology and medium shall be changed out
  - L-9w for turnover of unpowered Bioculture System
  - Use of LSG for crew operations
  - Standard health and status data, and non-standard data - cabin temperature, total pressure, relative humidity, ppCO2, ppO2
  - Use of three SSPF labs at KSC pre-launch

[1] = Exception 53146-0004, Bioculture On-Orbit Transfer Power Exception

Integration Overview (Continued)

- **Pre-flight specimen and hardware processing in the KSC SSPF**
- **On-orbit operations**
  - Majority of crew procedures already developed and executed on-orbit for CS-V (SpX-13) and upcoming CS-02 (SpX-15)
  - Updates for new operations for CS-03 are in work with POIC cadre
  - Training Strategy: OBT
  - Real-time support at ARC: Telemetry monitoring and commanding
  - Requesting PD enablement and over-the-shoulder video during operations in LSG
- **Use of ISS facilities:**
  - ExPRESS Rack
  - Life Science Glovebox (LSG)
  - MELFI (and Cold Stowage on Dragon – Polar and/or DCB)
  - Wet Lab Pantry Items (nitrile gloves)
- **Asynchronous Ground Control at ARC**
- **Post-flight science recoveries at PI laboratories.**

Stowage Overview: Ascent on SpX-16

Unpowered Locker

- Bioculture System
  - Containing 10 Cassettes with no live cells

Cold Stowage

- Cell Loading Kit (x2)
  - -80°C or colder
  - Each contains five 3mL syringes containing frozen cell lines
- Media Stowage Bag (x9)
  - +4°C
  - Each Media Stowage Bag contains five Media Bags (total of 45)

Soft Stowage

- Bag Changeout Kit (x4)
  - Contains empty Sump Bags, Hemostats, Wipes, and other accessories
  - One kit contains supplies for 10 cassettes.
- Bioreactor Removal Kit (x1)
  - Contains Tube Cutter, two hard-sided Bioreactor Containers, and stowage bags
- Sample Bag Removal Kit (x1)
  - Tube cutter and Sample Stowage Bags
- Gas Supply (x3)
  - Replacement Gas supplies to be installed during gas supply changeout
- Setup Kit (x1)
  - Items for setting up LSG (wipes, O-rings)
- Accessories Kit (x1)
  - Spare syringes, hemostats, caps, wipes, etc.

Stowage Overview: Descent on SpX-16

Cold Stowage

- Bioreactor Container (x2)
  - -80°C
  - Each container will have five individually bagged Bioreactors
- Sample Stowage Bag (TBD)
  - -80°C
- Return Bag (TBD)
  - -80°C (Media Bags)
- Return Bag (TBD)
  - -80°C (Sump Bags)

Soft Stowage

- Bioculture System
  - Will be unpowered, removed from ExPRESS Rack, and packed in foam following conclusion of the experiment.
  - Will require oversize bag
  - Needs to be early retrieval at Long Beach
- Gas Supply (x3)
  - Used gas supplies for refurbishment

Backup Slides

CS-03 Hypothesis and Objectives

Hypothesis: Based on PI's previous results, CDKN1a/p21 may be a key molecular mechanism in the control of stem cell based tissue regeneration and is therefore a key candidate for stem cell-based tissue regenerative therapies and investigation in microgravity. This study hypothesizes that CDKN1a/p21 inhibits the proliferation and differentiation of mesenchymal stem cells into bone forming osteoblasts in space environment. Therefore, bone marrow mesenchymal stem cells from the CDKN1a/p21-null mice are expected to show unrestrained proliferation and differentiation in microgravity.

Objectives:

- **Specific Aim 1:** Assess the in-vitro proliferation, differentiation, and mineralization capacity of bone marrow mesenchymal stem cells isolated from CDKN1a/p21-null mice compared to wild-type animals in microgravity versus 1g controls.
- **Specific Aim 2:** Determine cellular mechanisms associated with alterations in osteoprogenitor differentiation potential in CDKN1a/p21-null mice versus wild-type controls
- **Specific Aim 3:** Investigate the signal transduction pathways, specifically NFκB, MAPK, and Pi3K signaling, which are responsible for activation of CDKN1a/p21 in microgravity and therefore inhibition of in vitro bone formation in space.