National Aeronautics and Space Administration


Credit: NASA

BIOCULTURE SYSTEM

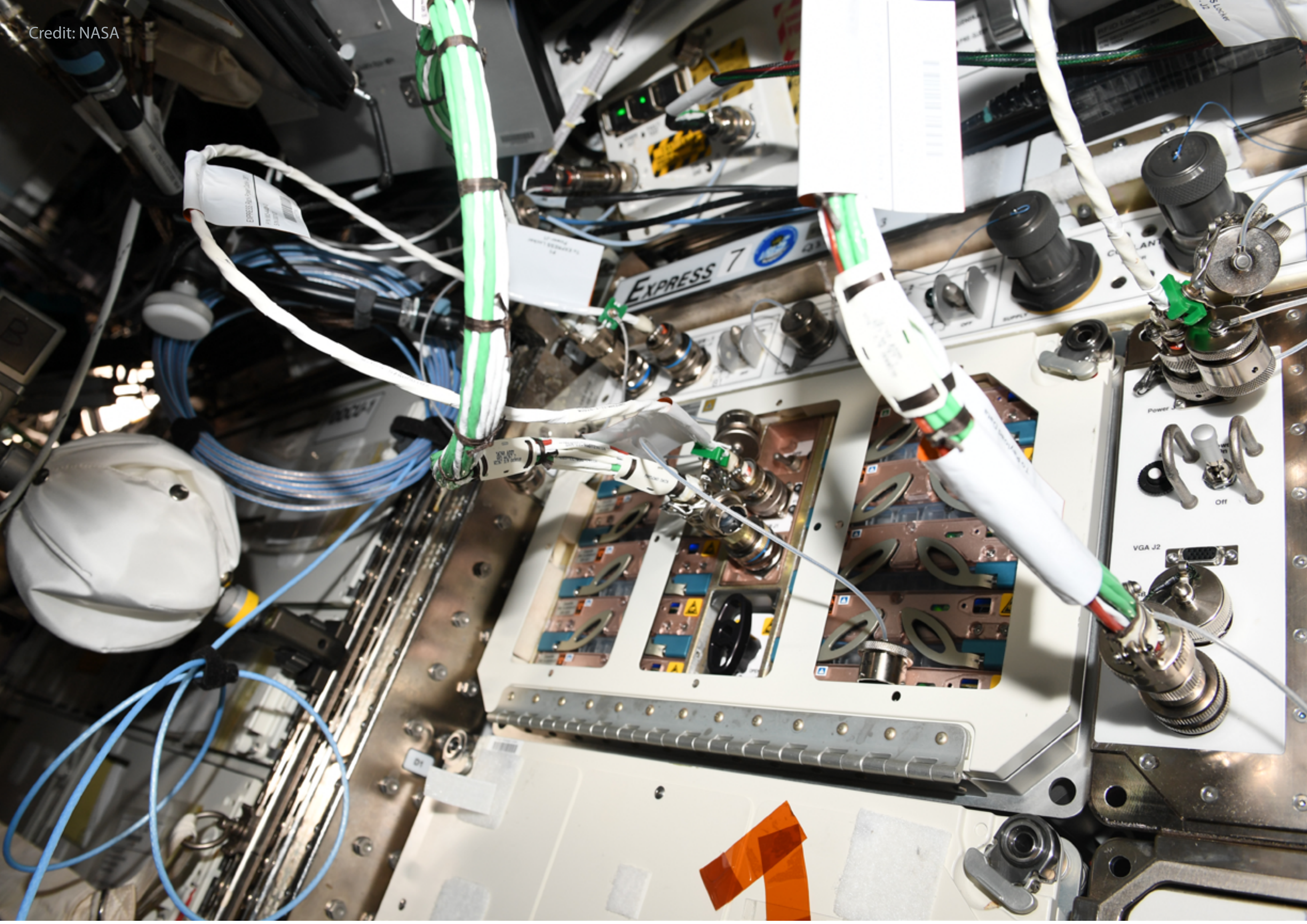
A Portable Biological Science Lab in Space

The Bioculture System is an automated and crew-accessible biological science incubator that provides controlled maintenance and growth of biology in a microgravity environment. Designed for use on the International Space Station (ISS) to support a wide diversity of tissue, cell, and microbiological cultures and experiment methods, this advanced system enables investigations on the ISS to address critical cellular and microbiological questions in space and on Earth.

The Bioculture System addresses a critical need for ISS National Lab academic and commercial space bioscience research by providing a platform system that can be reconfigured and customized to meet specific scientific objectives—a containment system that allows for initiation, intervention, and analysis of experiments on-orbit.

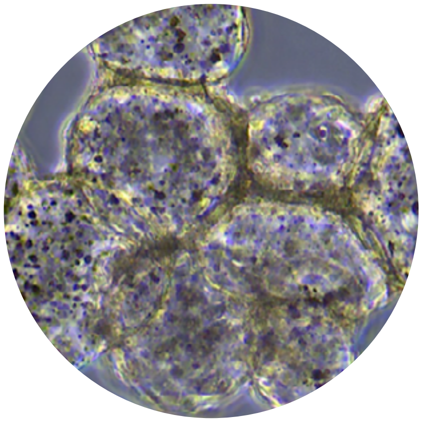
The Bioculture System incorporates ten independently controlled experiment cassettes, each with an independent perfusion-based cell culture bioreactor and computer-controlled fluidics to allow for multiple automated experiment manipulations. The automation includes standard laboratory culture maintenance such as temperature control, O_2/CO_2 delivery, and media change out. The system can also be programmed to include automated injections and fraction collections. The integral refrigerator compartment found in the cassette provides a critical capability to the system—extending and preserving the life of heat-labile components.

Credit: NASA



Credit: NASA

Beyond the automated features, the cassettes are designed for use in a biological safety cabinet (glovebox on the ISS), allowing for real-time culture initiation from frozen stores and for recovery of cells and samples. Investigators have the flexibility of utilizing nearly any cell or microorganismal culture and microbiology system of interest, which allows for a wide diversity of cell biology, microbiology, discovery biology, and drug-testing studies to be conducted on the ISS.



Credit: Natalya Dvorochkin

Credit: Christina Lim

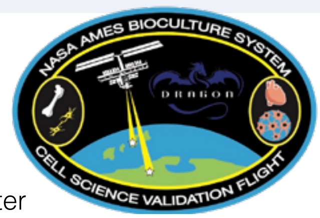
Credit: Miki Huynh

Research that improves human life, in space and on Earth.

History

The Bioculture System design concept and requirements were developed by NASA Ames Research Center (ARC), and initially engineered by Tissue Genesis, Inc. (TGI), to be the next generation of the Cell Culture Module (CCM). It incorporates flight-proven design and lessons learned from the extensive CCM flight series that flew aboard eighteen Space Shuttle missions from 1992 to 2011. In 2015, the Bioculture System was transitioned to ARC located in Moffett Field, California, where it underwent further development. The system was successfully validated on the ISS in December 2017 using mouse MLO-Y4 cells and human iPS-derived cardiomyocytes during the SpaceX-13 Cell Science-Validation mission (CS-V). The Bioculture System was next launched on SpaceX-18 (July 2019) supporting the Cell Science-02 (CS-02) experiment that investigated the effect of bone stimulating factors on proliferation and differentiation of mouse osteoblasts under microgravity conditions. In June 2021, tardigrades were cultured on the ISS in the Bioculture System in the SpaceX-22 Cell Science-04 (CS-04) experiment to characterize the genes involved in tardigrades' ability to change and survive in the confines of space. With each mission, the Bioculture System's capabilities were expanded to meet requirements and challenges of maintaining various cultures in the spaceflight environment.

Flown Missions



SpaceX-13: Cell Science-Validation (CS-V), NASA Ames Research Center

SpaceX-18: Cell Science-02 (CS-02), Dr. Rasha Hammamieh, USACEHR, DoD Space Test Program-Houston and Dr. Melissa Kacena (Co-I), Indiana University School of Medicine

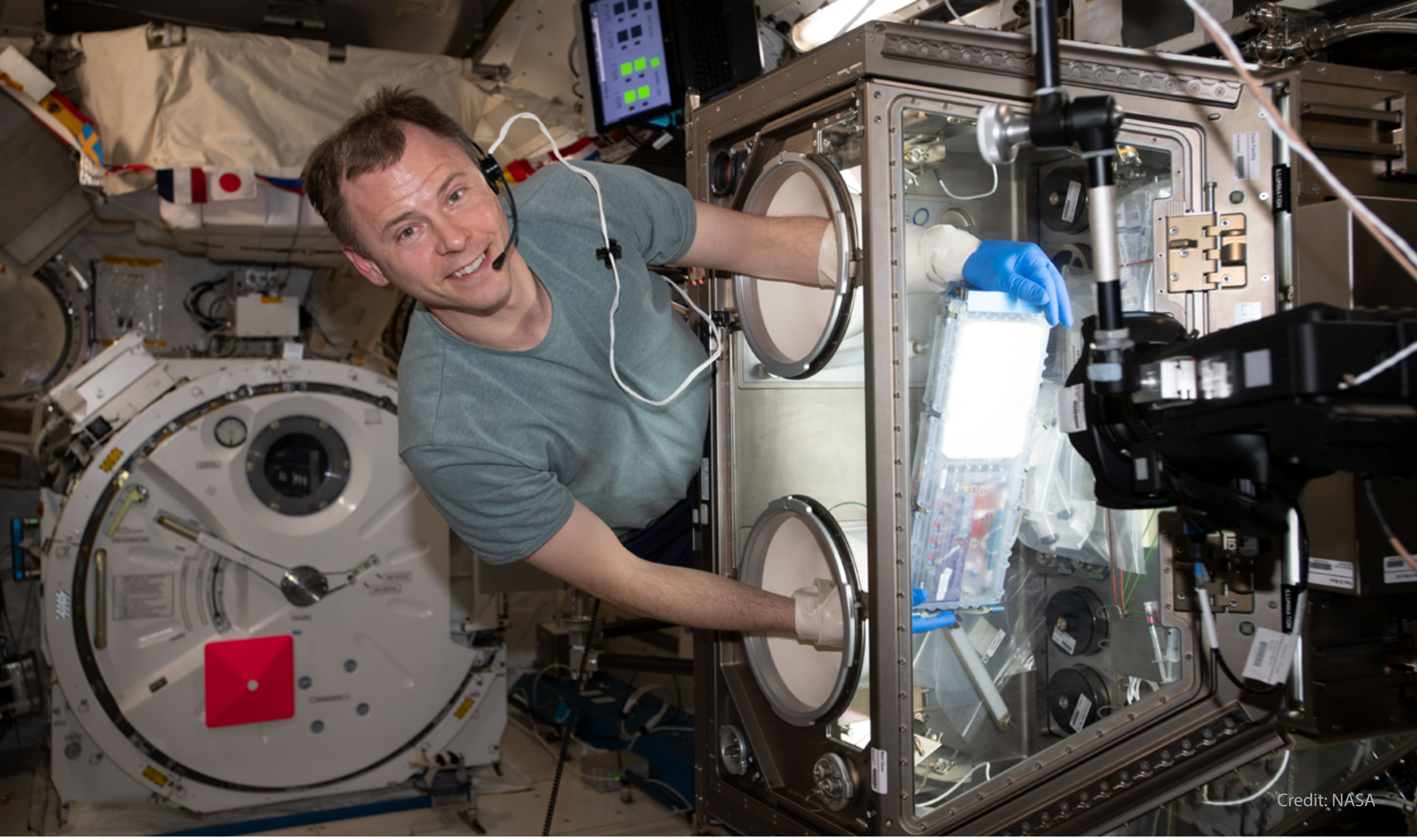
Space X-22: Cell Science-04 (CS-04), Dr. Thomas Boothby, University of Wyoming

Applications for the Future

Example Studies

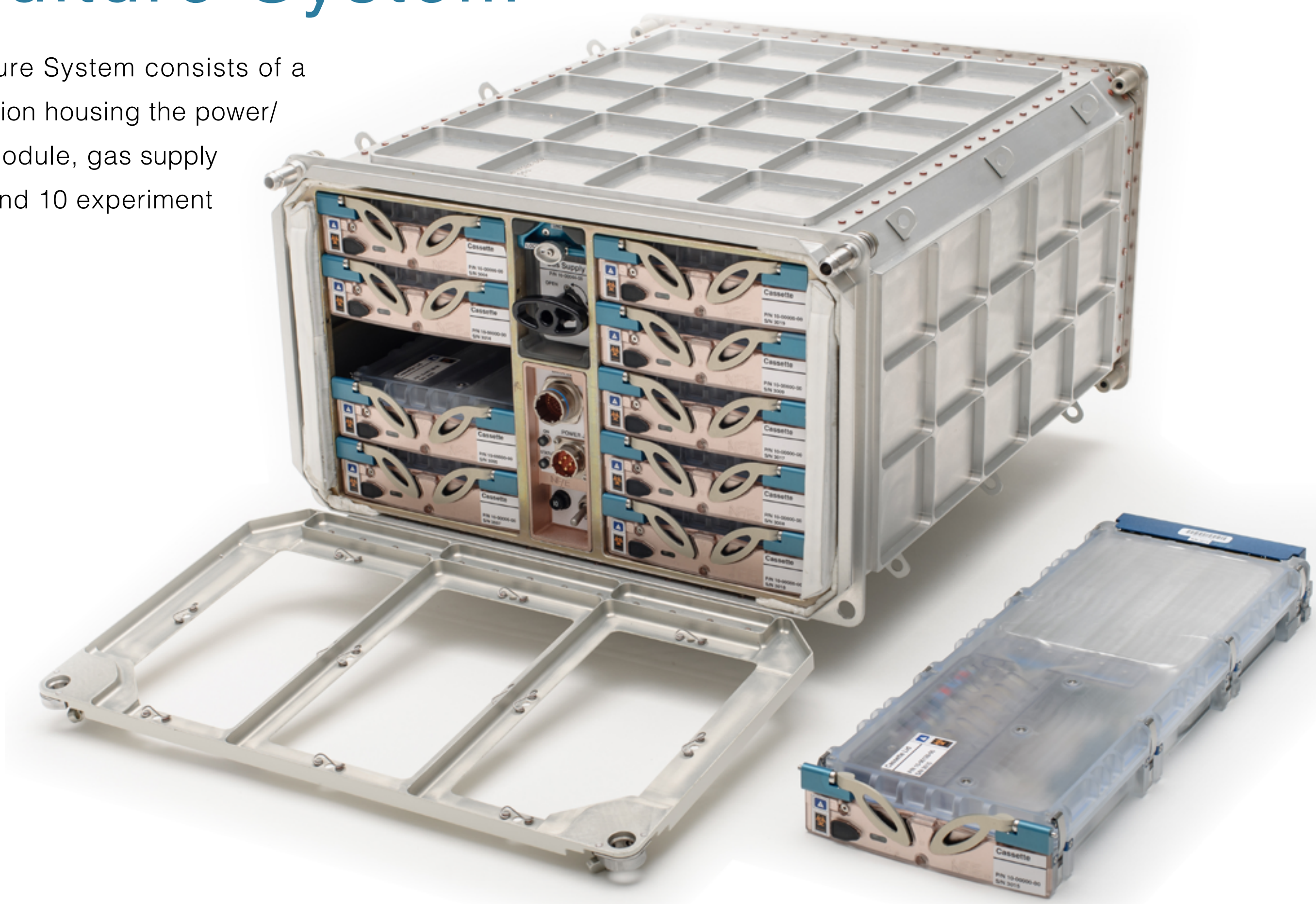
- **Basic cell physiology**
- **Cell cycle**
- **Genetics and gene expression**
- **Cell differentiation**
- **3D cell culture**
- **Tissue biology**
- **Immune cell function**
- **Cancer-related, radiation, drug discovery, commercial pharmaceutical discovery**
- **Drug compounds and countermeasure analysis and testing**
- **Basic microbe physiology**
- **Microbial virulence**
- **Biofilm research**
- **Host pathogen (bacteria and viruses) interactions**

Credit: Eye of Science


Credit: NASA

Bioculture System

The Bioculture System consists of a docking station housing the power/command module, gas supply assembly, and 10 experiment cassettes.


Credit: NASA/Dominic Hart

Cassette

The system allows the crew to remove a cassette from the docking station and transport it to the Life Sciences Glovebox (LSG) to access the fluidics/bioreactor and manipulate the experiment as needed to meet the science requirements. The cassette provides the structural support, power, data, gas supply, incubator and refrigerator compartments, and second and third levels of containments for the disposable flow path inside.

Credit: NASA/Dominic Hart

The closed flow path acts as a first level of containment for biologicals and hazardous materials that may be found within. The flow path configuration defines the organization of all pumps, valves, connections, bags, and bioreactors required for an experiment. The configuration of each flow path can be customized based on the requirements of the experimental plan.

Credit: NASA

The flow path is a one-piece, sterile barrier flow path with integrated bioreactor, pump, oxygenator, valves, preheat reservoir, media bag, sump bag, and fixative bag. The flow path also contains boards to drive the pumps and valves. These boards house an SD card that logs key experiment-related information such as temperature data, power draw data, valve states etc., as well as holds experiment command files (task file) for the flow path. Each flow path can run independent experiments from the others based on what commands are included in the task file. Possible manipulations include, but are not limited to, removing components of the flow path (e.g., sample bags), performing injections into the flow path, and taking samples from the bioreactor.

Credit: NASA/Dominic Hart

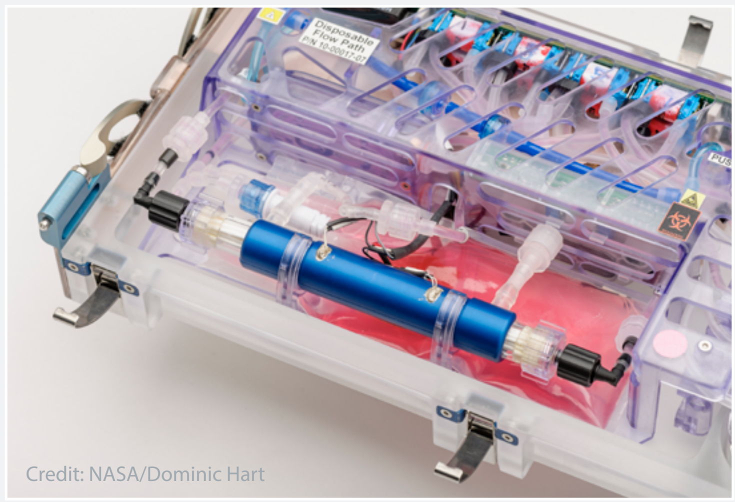
The Bioculture System can include standard laboratory preservative solutions (e.g. RNAlater or RNAprotect), maintained in the refrigerator compartment of the cassette. The solutions are automatically delivered into the bioreactor and mixed via the flow/reverse and trans-membrane flow capabilities of the system.

Hollow Fiber Bioreactor

The Bioculture System is designed to emulate perfusion culture systems such as CellMax Quad (Cellco) and FiberCell Duet (FiberCell).

The unique properties of the hollow fiber bioreactor make it an ideal choice for experiments conducted in microgravity where nutrient transfer is limited to diffusion. The multi-chambered, space-efficient design provides the best environment for successful cell culture. The bioreactor contains porous cylindrical hollow fibers embedded in the bioreactor shell in a parallel array. The culture medium flows inside of the fiber volume called the Intracapillary Space (ICS) and cells are cultured on the outside and around the fibers in the space called Extracapillary Space (ECS). This framework allows the fibers to create a defined semi-permeable barrier between the growing cells and the flowing medium, protecting cells from the fluid shear stress. It also allows for a continuous exchange of waste for nutrients as nutrients and oxygen diffuse into the ECS compartment through the porous fibers, while the metabolic wastes and CO_2 diffuse out and into the ICS, unlike in a typical lab setting where the waste and nutrients must be monitored and exchanged manually. This design allows efficient nutrient delivery, improving viability and reducing apoptosis.

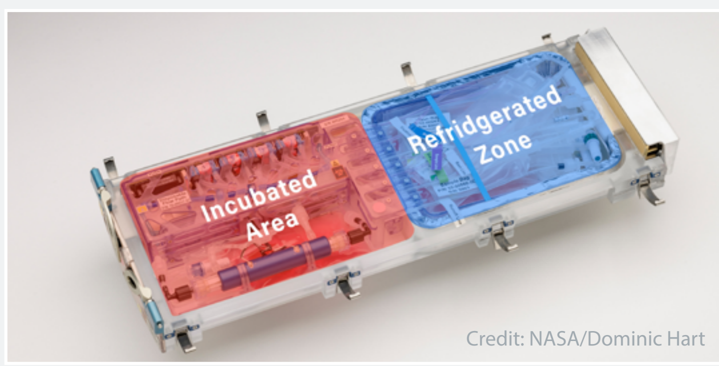
Credit: NASA/Dominic Hart



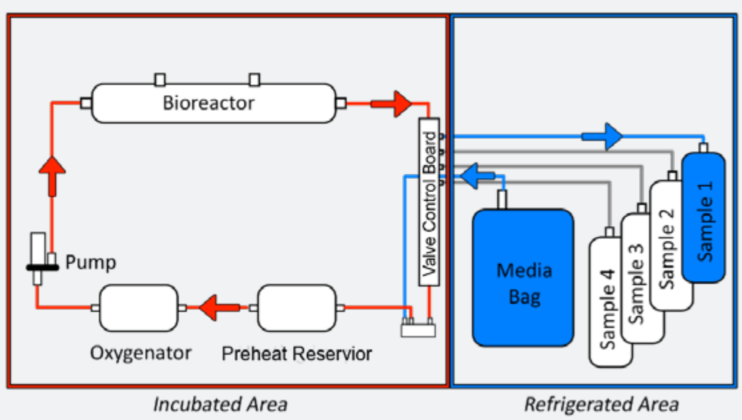
Credit: NASA/Dominic Hart

Incubation/Refrigeration

In most cases, the Bioculture System is programmed to maintain a constant 37°C environment. However, set point of the incubator side of the cassette can be adjusted from ambient temperature to 42°C. Each cassette includes a 200ml refrigerator compartment meant to store heat-labile media, additives (growth factors, antibiotics, etc.) and sample collections. The temperature set point of the cold compartment can be programmed from 5°C to ambient. Also, the temperature set points of the incubator and refrigerator sides can be modified near real time via ground commanding. The 200ml cooling chamber may be used to house the experiment biology where reduced temperature quiescence is desired for such models as embryos, bacteria, or other reduced metabolic protocols.

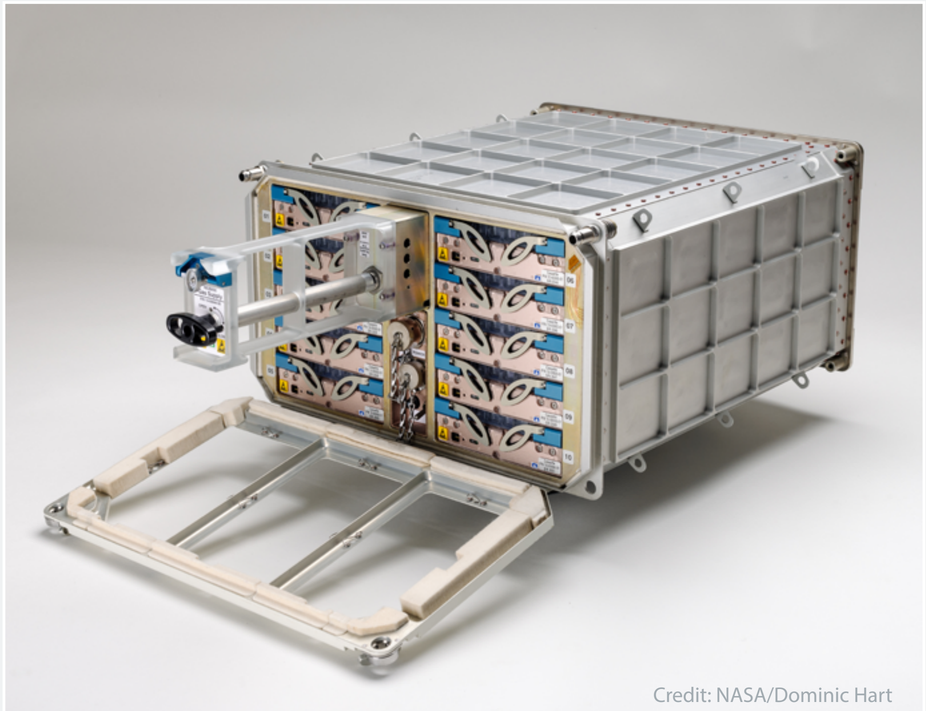


Credit: NASA/Dominic Hart

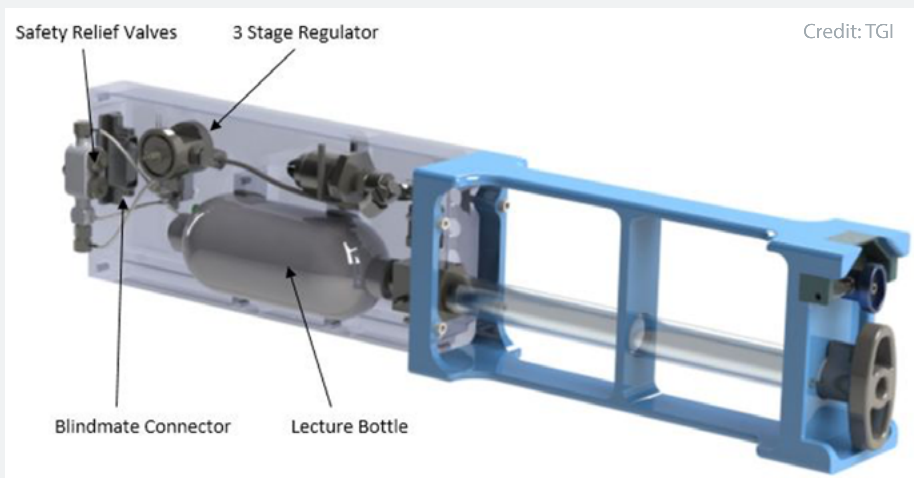


Gas Exchange

The Bioculture System can be outfitted with its own gas supply typically consisting of 20% O_2, 5-10% CO_2, and balanced with N_2. The gas supply assembly is integrated into the Bioculture System Docking Station and contains a 150cc gas lecture bottle with a three-stage pressure regulator to reduce the working pressure of 1800 psi to 4 psi. The assembly is removable/replaceable during on-orbit operations. Customizable gas mixtures, including a no gas option, can be designed to meet the research of interest. The gas is delivered through a manifold to a valve in each cassette. Each flow path contains the oxygenator that employs a series of silicone flow loops to provide gas exchange across the semipermeable tubing into the circulated culture medium. The sealed oxygenator has a one-way gas relief valve that prevents outside gases from entering in. The gas is supplied at a specified rate which can be adjusted during the experiment via ground commanding.

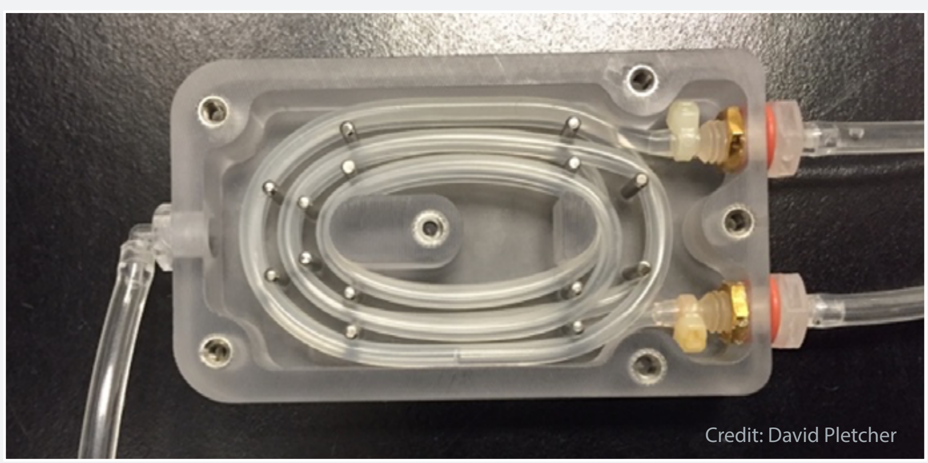


Credit: NASA/Dominic Hart



Credit: TGI

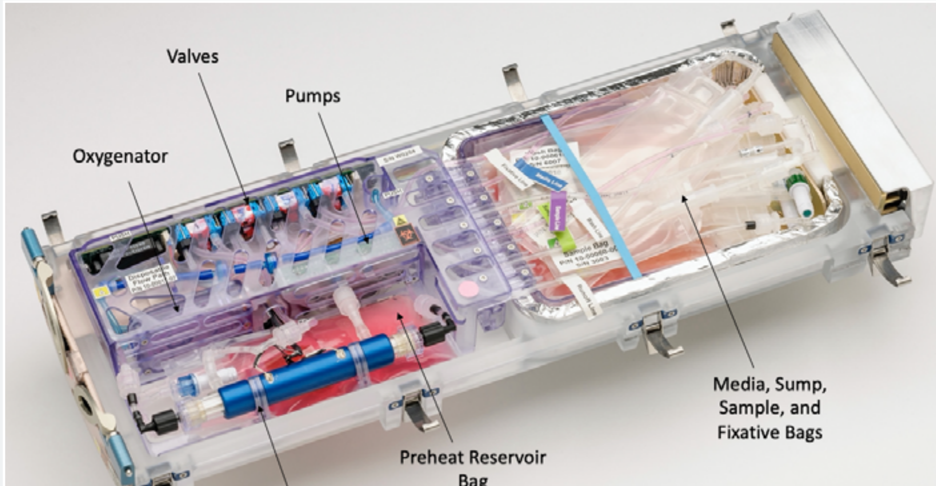
Credit: David Pletcher



Credit: David Pletcher

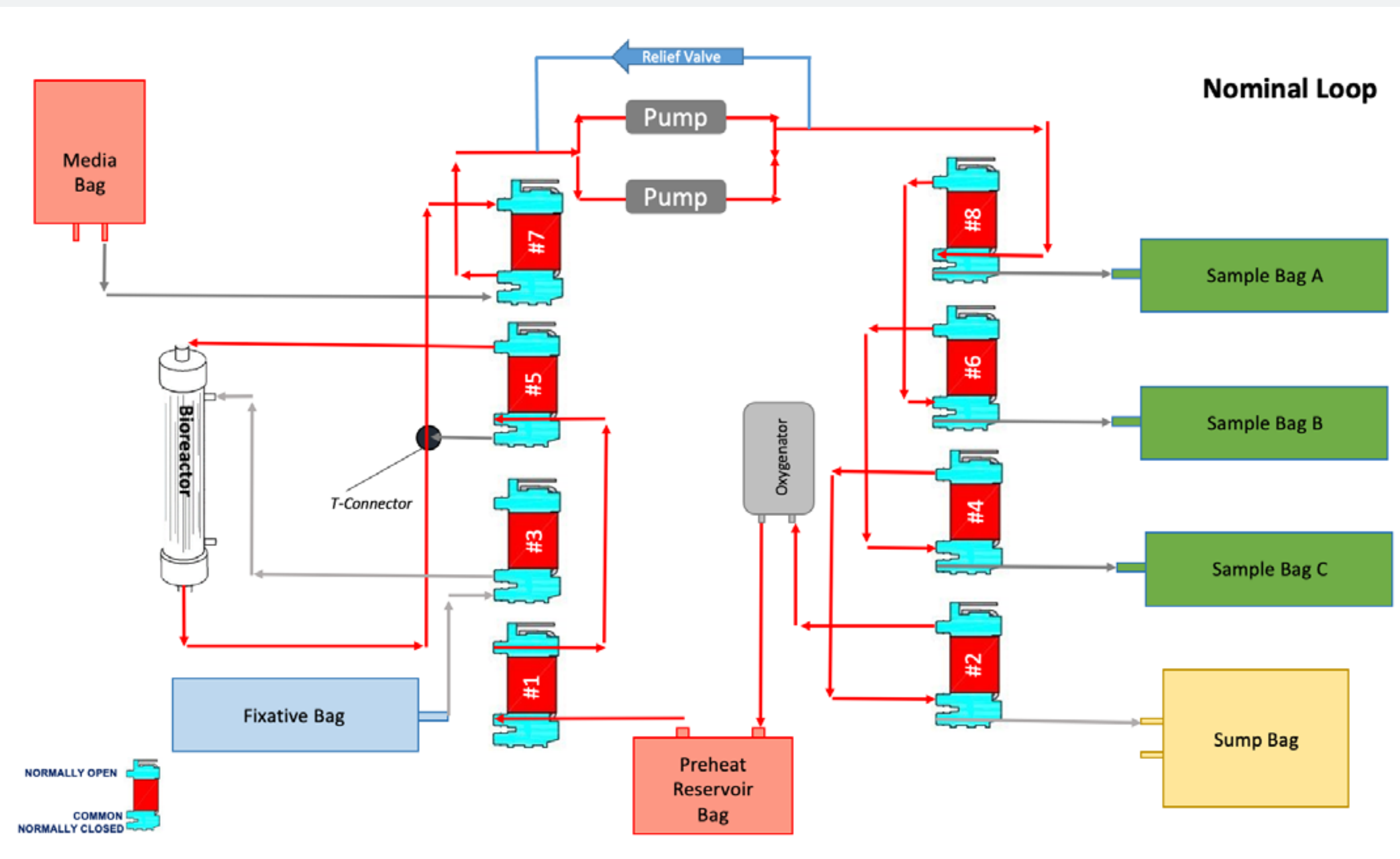
Media Delivery

The media delivery system, which features a closed loop flow path design, provides the ability to perform most cell maintenance operations and science manipulations. The continuous flow of medium through the bioreactor supplies cells with nutrients and dissolved oxygen, and CO_2 for pH maintenance, and allows introduction of other fluid reagents such as growth factors, inhibitors, and fixatives. The flow path fluidics routing is customizable for the experiment, but it generally includes two nominal nutrient delivery paths.



Credit: NASA/Dominic Hart

The first path is nominal recirculation of media through the preheat reservoir bag, oxygenator, pump, and bioreactor. In this fluidics route, approximately 60ml of media is continuously circulated through the bioreactor to exchange waste for nutrients. The second path is an intermittent feed which periodically pumps spent medium from the preheat reservoir into a waste bag and then pumps fresh medium from the media bag into the preheat reservoir. If cold, this fresh media can be recirculated in a bypass loop that allows media to reach the experimental temperature range before exposure to cells in the bioreactor. In addition to the two nominal fluidics routes, injections and samples can occur from either the refrigerator or incubator side of the flow path.



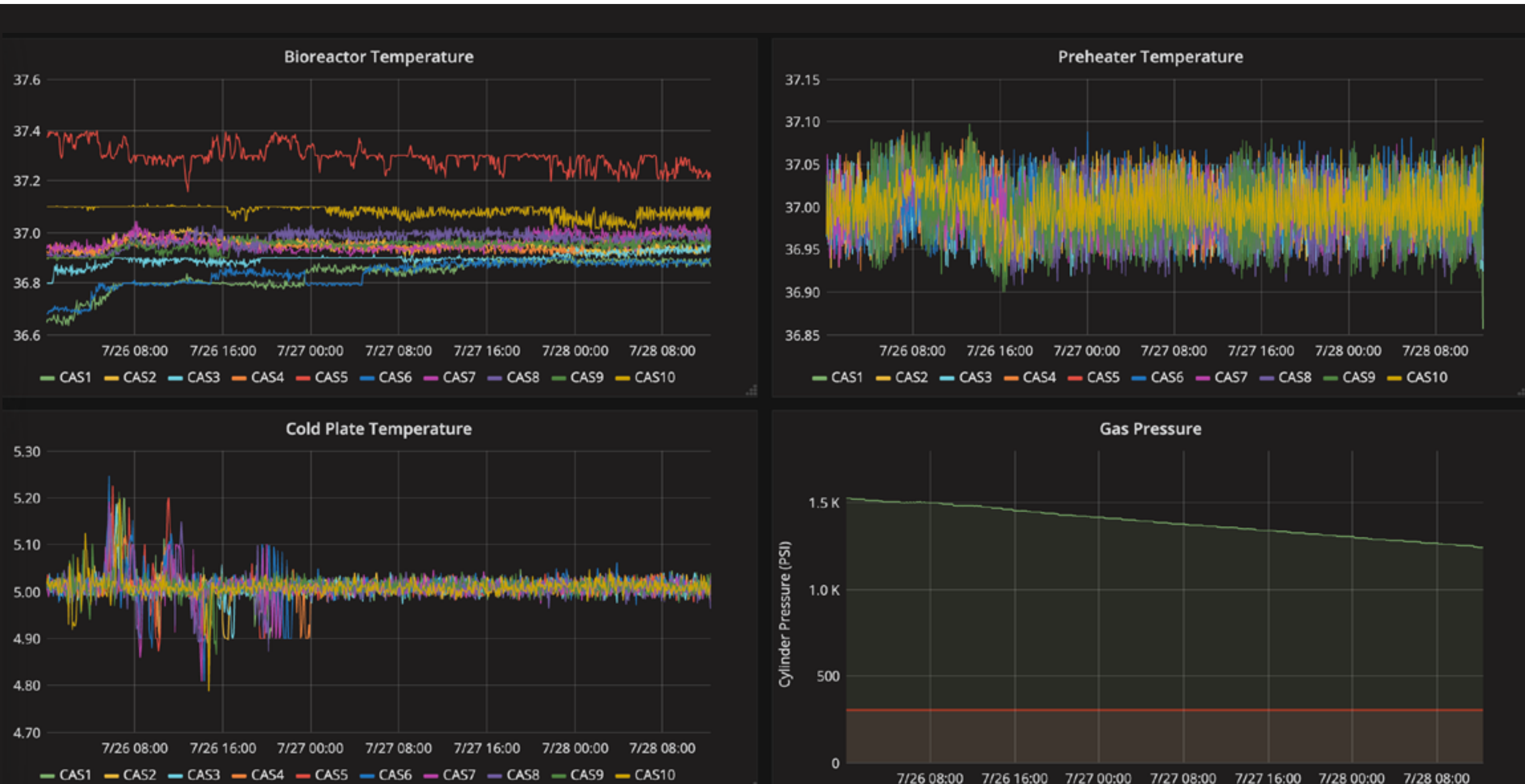
Real-Time Monitoring and Ground Control Access

Using a custom control system, the experiment is monitored and commanded from a ground site. This allows the investigator to receive real-time status updates and enables near real-time modification of experiment protocol set-points and automated events. Event schedule modifications may be dictated by launch, initiation, or astronaut timeline constraints. Examples of commanding performed through task files uploaded either automatically (pre-programmed prior to launch) or by ground commanding are automated control of cassette environment and sensors, automated fluid exchange, hardware commanding for temperature changes, and data downlink.

Examples of critical parameters monitored from the ground:

- ◊ Cassette temperatures at bioreactor, preheat reservoir bag, and cold plate
- ◊ Bioculture System temperatures at inlet and outlet, control board, Power and Control Module (PCM), and Thermo-electric Cooler (TEC) heatsink
- ◊ Gas pressure
- ◊ Current
- ◊ Power cycle

Console operations are performed from the NASA ARC Multi-Mission Operations Center (MMOC) at Moffett Field, CA. Crew operations in the Life Sciences Glovebox (LSG) on the ISS have an assigned member of the Payload Developer team "in the ear" of the crew performing the operations. Staffing is critical for successful operations, and include Operations, Software, Engineering, Science, Project Management, and a PI representative.




Credit: NASA/Dominic Hart


Credit: NASA

Experiment Design and Development

Typical Experiment Design Example

A typical Bioculture System experiment utilizes a set or series of pre-programmed events to accomplish experiment objectives. An experiment can combine periodic on-orbit media exchange with media sampling and fixation of specimens. A typical study might examine the effects of in-flight hormone stimulation by collecting media fractions at pre-set intervals post-exposure. Injection of treatment can be automated. Crew operations may include initiation of culture on-orbit and specimen transfer to ISS cold stowage assets for long duration stowage.

Top Level Experiment Timeline

Investigators should expect to spend one year conducting ground studies and science/hardware compatibility testing prior to flight. However, the actual duration (longer or shorter) for ground studies and tests will depend on the complexity of the experiment, cell type, and if the specimen was previously flown or grown in a hollow fiber system. Once selected for flight, compatibility with the Bioculture System is assessed and hardware and science requirements are defined. Six months to a year may then be spent adapting the experiment to the Bioreactor System and optimizing growth conditions. When the science and hardware parameters are selected, a full scale mission duration test may be performed using the fully flight configured Bioculture System at an appropriate test facility (e.g. NASA ARC or NASA KSC). The mission duration test aids both scientists and engineers in mission and contingency preparation, including final biocompatibility tests, ground baseline experiment data collection, sterile processing concerns, procedures, pre-flight timelines, mission delays/scrubs, and the logistical concerns of “taking an experiment on the road.”

Reference:

NASA Bioculture System:
https://www.nasa.gov/ames/research/space-biosciences/bioculture-system

Investigators interested in using the Bioculture System for experimentation please contact:

Robert Vik
robert.d.vik@nasa.gov

Bioculture System Specifications

Physical

- 72lb control weight
- Late access payload (24 hours prior to launch)
- Triple containment of biological and hazardous chemicals
- Power: 150 Watts max
- Synchronous or asynchronous flight and ground controls

Media Delivery

- Individual flow paths (biocompatible components such as Flexelene 75E, silicone, polypropylene, nylon, etc.)
- Feeding Schedules: continuous recirculation and preprogrammed feeding schedule based on experiment needs
- Gas delivery and pH maintenance

Drug/Fixative Delivery System

- Programmed delivery schedule
- Cooled heat-labile drugs and fixatives
- Programmed injection of drugs or fixatives to the system

Environmental Control

- Module temperature: Ambient to +42°C±1.0°C
- Cooling chamber temperature: Ambient to +5°C±1.0°C

Gas Delivery System

- Computer controlled, high pressure solenoid distribution
- Typically, medical grade gas mixture: 5-10% CO_2, 20% O_2, balanced with Nitrogen (for CO_2-dependent media), or breathing air (for organisms/cells not requiring CO_2)
- Membrane oxygenator using silicone elastomer tubing
- Positive pressure airflow within the system
- Programmable delivery schedule

Bioreactors

- Hollow fiber bioreactor or custom-design bioreactors
- Sustains attached and non-attached cell types with or without microcarrier beads
- Variable molecular weight cutoff

Fluid System

Sustains a maximum of 10 independent flow paths.

Each fluid path may consist of the following:

- Media bag
- Sample bags
- Solenoid valves
- Pump (0-8 mL/min flow rate)
- Membrane oxygenator
- Space flight qualified hollow fiber bioreactorcartridges or alternate selected by investigator and verified in system
- Biocompatible tubing that can accommodate hazardous fluids (fixatives, etc.)
- Programmed injection of drugs or fixatives to the system
- Fraction collections (10/flow path)

System Redundancy

- Individual flow paths
- Individual experiment cassettes
- Individual flow rates and pump control
- Individual media and gas feeding schedules
- Independent experiment program and operational control
- Independent temperature control

Electronics

- Power driver board for each cassette
- Dedicated controller and data logger for each cassette