



### FFL – 01 Points of Contact

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### Hardware Partners Points of Contact

- Observation System Developer:
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- Controller Unit Developer:
   Luca Pieroni, Kayser Italia, <u>I.pieroni@kayser.it</u>
- Software Developer: Arshad Mian, Intrinsyx, arshad@intrinsyx.com
- NanoRacks Facility:
   Chris Cummins, NanoRacks, ckcummins@nanoracks.com



#### **Description and Objectives:**

- Theme: (Decadal Survey Area): AH14, AH16, CC2, CC8, CC10
- Description: To support multi-generational experiments with Drosophila melanogaster (fruit flies) at various gravity levels (0 to 2 g).
- **Implementation**: Two phase approach to provide an immediate capability using existing hardware, and then to develop new fly hardware that meets the full requirements.
  - Phase I Use FIT Fly cassettes in the Nanorack Centrifuge
  - Phase II Develop Fly EUE for either the EMCS or TechShot Multi-specimen Variable-g Facility (MVF) with environmental control, added containment, and fixation.
- Schedule:

Validation FFL-1 SpaceX-5, FFL-2 SpaceX-7, Yearly flights planned

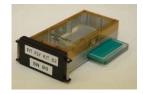
#### Approach:

A small diameter centrifuge onboard ISS will provide the capability to study the flies in partial-g and 1-g. The NanoRacks BioRack Centrifuge and µg-Rack will accommodate FIT fly cassettes inside a Type-1 container. Video imaging is currently being developed. Fixation capability under consideration.



Drosophila melanogaster (fruit fly)

Phase 1 HW



Possible Phase 2 HW



#### **Justification:**

Value to Agency (Space Benefit)

This system enables studies of genetic responses to microand fractional-gravity and effects on reproduction in a complex organism that has been extensively used in labs around the world for such studies. This is a capability that is lacking, but desired, by all of the international partners for onorbit space biology research.

• Value to Public (Earth Benefit)

Microgravity exposure has unmasked genetic mechanisms in simpler organisms, and needs to be studied in more complex organisms. Strong potential for education outreach paired with the science



## Heritage

• **Aim:** Replicate and extend scientific return from FIT experiment, STS 121, 2006 (PI for FIT mission: Dr. Sharmila Bhattacharya, NASA ARC) and baseline future Drosophila spaceflight hardware capabilities.

### Science data obtained from FIT:

- · Reduced immune function after spaceflight
- Developmental changes & slight increase in lethality (presumably at pupal stage)
- All assays conducted immediately after landing with live samples (genomics, phagocytosis cellular assays, clearance of gram negative E.coli infection postflight, counts of animals in each developmental stage to assess increased lethality)
- Data published in 2011 (*PLoS One*; 6(1): p. e15361, doi:10.1371/journal.pone.0015361)

#### Science Data to be Obtained on FFL-01:

- Immunological, developmental, cellular, and molecular changes and lethality assessment following bacterial challenge in-flight assessed via:
  - Imaging of insects during flight
  - · Dissection of and examination of insects post-flight
  - Protein/gene expression during and post-flight (freeze/fix)



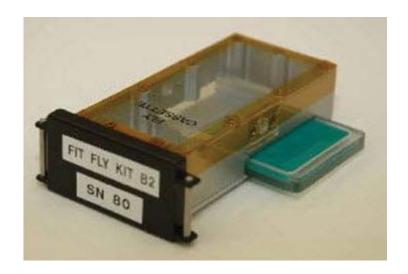
# Validation Objectives

- Support Drosophila studies on ISS by upgrading capabilities of existing hardware that supported successful immunology study on STS-121.
- Demonstrate reflight of FIT hardware on existing NanoRacks centrifuge facility to provide on-board 1g control experiments for the first time.
- Utilize freezing and/or fixation to overcome increased sample handover time (compared to previous shuttle flights) thus demonstrating the science utility of Space-X flights.
- Implement fly treatment technique to study immune response of organism to inflight challenge.
- Gather behavioral data using a new camera system compatible with existing microgravity and centrifuge hardware.
- Demonstrate ability of hardware to support multigenerational studies on-orbit.

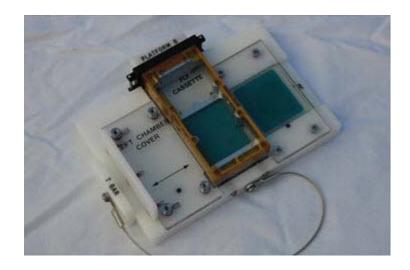


## **Experiment Hardware**

The FFL hardware suite consists of fly cassettes and platforms used for food change out that were successfully flown on STS-121 in 2006 during the FIT project.



Fly Cassette with food tray partially inserted.

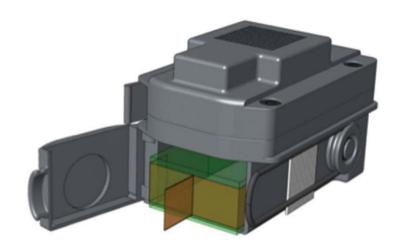


Changeout Platform with cassette. Food tray in right slot. **Tethered T-bar** inserted in left slot.

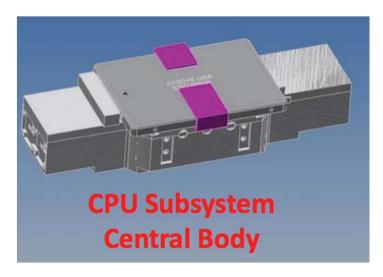


## **Experiment Hardware**

- An observation system is being developed by Astrium, Kayser Italia, and Intrinsyx based on the form factor of a standard ESA Type-I container
- Each fly cassette will slide into an observation unit that will have a camera, LED's for day/night light cycling, and infrared LED's to enable recording during dark cycle.
- Cameras will be cycled by controller units and data will be stored on incorporated SSD's.



Observation Unit with fly cassette installed.

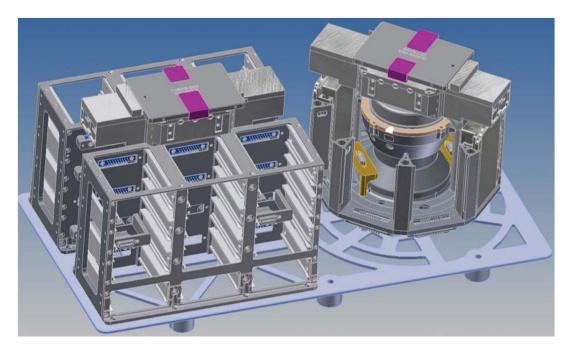


Controller Unit with SSD installed.



# NanoRacks Facility

- Observation units, all containing fly cassettes, will be inserted into slots on the BioRack Centrifuge and μG-Rack located in NanoRacks Platform 3.
- 6 units each on the  $\mu$ G-Rack and BioRack centrifuge for on-board 1g control (12 units total).
- Controller units and associated SSD's will be placed on top of each rack (2 total).

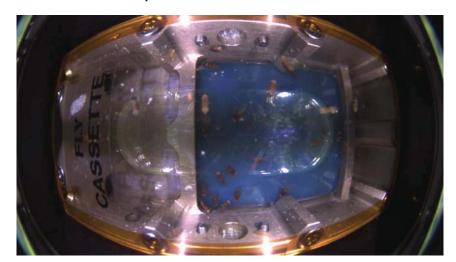


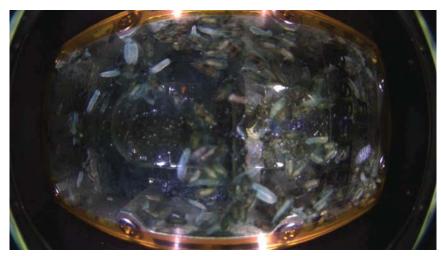
NanoRacks BioRack centrifuge and µG-Rack with controllers installed.



# **Observation System Data**

- Camera units will collect video data throughout mission and store data onto SSD's.
- SSD's will be swapped out throughout flight, likely during food changeout operations, and stowed for return.
- For in-flight fly culture health check, short video clips or still images may be captured intermittently and transferred via Eye-Fi SD Wi-Fi card to on board computer or ISS Wi-Fi for downlink.





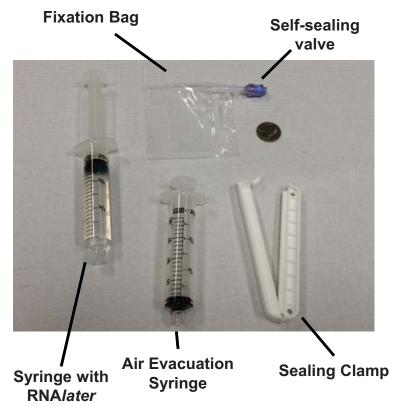
Day 0 → Day 24

Stills generated by prototype camera unit showing development of fly cultures in cassettes over 3+ weeks.



### **Fixation Hardware**

- Fixation is likely to be added for FFL-01.
- Inside WetLab Disposable Glove Bag (DSB), food tray will be removed from cassette using changeout platforms, and inserted into BioServe Fixation Bag.
- RNAlater will be inserted with BioServe Kit syringe, and bags will be frozen in MELFI for return.





# **Pre-flight Operations**

- Twelve fly cassettes will carry initial generation of flies.
- Organisms will be flown from ARC to KSC 2 weeks prior to launch to begin growth of flight cultures.
- Day of handover operations:
  - Prepare Drosophila food and pour into food trays.
  - Add microbial organism to subset of food trays to be used for onboard treatment.
  - Install all food trays in initial 12 cassettes or change-out platforms.
  - Transfer initial generation of adult flies to 12 cassettes.
  - Package all hardware.
  - Handover.
  - Continue collecting appropriately aged flies for scrub turnaround replacement.

# **On-orbit Operations**

Op #1

Launch

FD ~1 Install 6 cassettes containing flies each into observation units and onto centrifuge and µg rack. Install controllers and SSD's and begin experiment.

Op #2

FD ~5/6 new food tray provided to old cassettes. Old food trays frozen in new cassettes.

Op #3

FD ~12 old food trays transferred to new cassettes. Old cassettes frozen. 3 cassettes processed for fixation in DSB.

Op #4

FD ~20 treated food trays provided to ½ old cassettes. Old food trays frozen in new cassettes.

Op #5

FD ~26 subset of old food transferred to new cassettes. Both frozen. Two cassettes given new food for live sample return.

Op #6

FD ~28 Disassemble hardware and pack for return. All frozen cassettes transferred to POLAR. Live samples packed for ambient return.

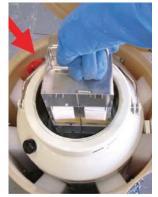


# Post-flight Operations

- Live samples will be collected at the port and visually assessed for health & viability by Project Scientist/Science Team.
- Live samples and ambient hardware will be driven to ARC for further processing.
- Frozen samples will remain under control of the Cold Stowage group to JSC.
- Member of payload team will receive frozen samples at JSC, package in cold nitrogen vapor dewar, and prepare for shipping to ARC.
- Samples will be received by science team at ARC for inspection and processing.
- Some hardware will be disassembled, cleaned, and reassembled to be used for ground control.









### **Ground Control**

- An asynchronous ground control will be performed at ARC after flight return.
- Environmental temperature and humidity data will be collected via a data logger throughout the flight.
- Ambient CO<sub>2</sub> and O<sub>2</sub> levels will be collected from ISS.
- Environmental parameters will be replicated by a programmable chmaber.
- Flight hardware including Observation System, change-out platforms, and some food trays will be reprocessed and used for the ground control.
- A separate set of fly cassettes will be used (flight set will be stored frozen for processing).



# **Crew Training**

- Training Strategy Team (TST) Conducted March 18, 2013.
- OBT selected for crew training, with introductory video and embedded short video clips in procedures.
- Ops are time critical to docking, will schedule OBT of introductory video a few days prior.
- Spare cassette and changeout platform will be flown to be used by crew for hands-on familiarization prior to performing experimental operations.
- Spare hardware will be left on ISS for future flights.

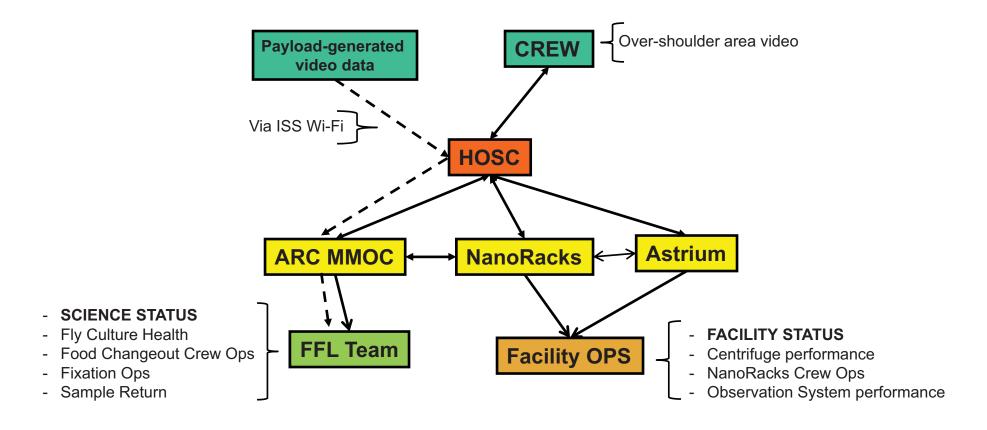


### **Crew Procedures**

- Food change-out procedures currently drafted.
- Revised from heritage procedures performed on same hardware flown during project FIT on STS-121.
- Necessity of using MWA currently being determined.
- Monthly video-conference run-throughs conducted with payload team, operations lead, crew office rep, PIM, PARC, and NanoRacks rep.
- Observation System installation and disassembly procedures beingn developed in cooperation with hardware developers.
- Fixation feasibility study complete and high level procedure developed using BioServe hardware and WetLab Disposable Glove Bag.



# Real-time Operations Interface





# Fruit Fly Lab – 02 (SpaceX-7)

"The effects of microgravity on cardiac function, structure and gene expression using the Drosophila model."

Principal Investigator:

Rolf Bodmer, Ph.D., Sanford Burnham Medical Research Instititue rolf@sbmri.org

### Aims:

- To quantify the response of the heart to microgravity in both male and female flies
- To quantify the response to microgravity of hearts from flies genetically predisposed to arrhythmia and cardiac dilation
- To identify cardiac gene expression changes in response to microgravity
- To determine the how long the effects of microgravity persist in the heart following return to earth gravity.



# Fruit Fly Lab – 02

### **Key Deltas:**

- Science mission utilizing technologies demonstrated in FFL-01.
- Fixation, if utilized, will be performed on experimental samples housed on BioRack.
- Relies heavily on post-flight analysis of live animals and will therefore have significantly more live samples returned and fewer frozen.
- Requires +12C stowage post-processing on ISS and during return on Dragon to slow development of samples for post-flight processing of specific ages.



# Back-up Slides



### **Crew Operations Detail**



### Op #1

#### 1 hour

- Remove from launch packaging
   12 cassettes containing flies.
- 2. Insert 6 cassettes into doublevolume type-1 containers on μg rack.
- Insert remaining 6 cassettes into double-volume containers on centrifuge.
- 4. Set centrifuge to Earth gravity and start centrifuge.
- 5. Initiate video recording (TBD).

### Op #2

#### 2 hours

- Retrieve from 4C cold stowage 12 changeout platforms containing food.
- 2. Remove 12 cassettes from µg rack and centrifuge.
- 3. Place cassette into center of changeout platform, aligning food trays in platform and cassette.
- 4. Using tethered T-bar, insert new food tray into cassette.
- 5. Remove cassette from platform and insert into double-volume type-1 container on µg rack or centrifuge.
- 6. Place new blank cassette into used platform and insert old food tray using tethered T-bar.
- 7. Place new cassette containing old food tray into MELFI.
- 8. Repeat for all 12 cassettes.
- 9. Restart centrifuge.
- 10. Initiate video recording (TBD).

### Op #3

#### 2 hours

- Retrieve from ambient stowage 12 changeout platforms containing blank food trays.
- 2. Remove 12 cassettes from µg rack and centrifuge.
- 3. Place cassette into center of changeout platform, aligning food trays in platform and cassette.
- 4. Using tethered T-bar, insert blank food tray into cassette.
- 5. Remove old cassette from platform and place into MELFI.
- 6. Place new blank cassette into used platform and insert old food tray using tethered T-bar.
- 7. Remove new cassette containing old food tray from platform and insert into double-volume type-1 container on µg rack or centrifuge.
- 8. Repeat for al 12 cassettes.
- 9. Restart centrifuge.
- 10. Initiate video recording (TBD).



# Crew Operations Detail (cont.)



### Op #4

#### 2 hours

- Retrieve from 4C cold stowage 6 changeout platforms containing food and 6 platforms containing food with treatment.
- 2. Remove 12 cassettes from µg rack and centrifuge.
- 3. Place cassette into center of changeout platform, aligning food trays in platform and cassette.
- 4. Using tethered T-bar, insert new food tray into cassette.
- Remove cassette from platform and insert into double-volume type-1 container on µg rack or centrifuge.
- 6. Place new blank cassette into used platform and insert old food tray using tethered T-bar.
- Place new cassette containing old food tray into MELFI.
- Repeat for all cassettes using 3 platforms with treated food each for µg and centrifuge cassette subsets.
- 9. Restart centrifuge.
- 10. Initiate video recording (TBD).

### Op #5

#### 2 hours

- Retrieve from ambient stowage 12 changeout platforms containing blank food trays (previously used in Op #3).
- 2. Remove TBD cassettes from μg rack and centrifuge.
- Place cassette into center of changeout platform, aligning food trays in platform and cassette.
- 4. Using tethered T-bar, insert blank food tray into cassette.
- 5. Remove old cassette from platform and place into MELFI.
- Place new blank cassette into used platform and insert old food tray using tethered T-bar.
- Remove new cassette containing old food tray from platform and place into MELFI.
- 8. Repeat for TBD cassettes.
- Stow remaining cassettes in launch packaging for live sample return.

### Op #6

#### 1 hour

- Transfer all hardware to appropriate stowage on Dragon.
- TBD µg rack and centrifuge shut down.



## **Hardware Stowage**



