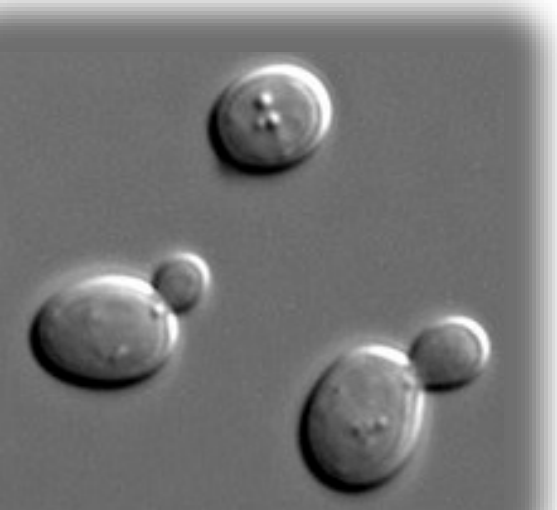
The Usefulness of Small Model Organisms in Spaceflight Research

Sharmila Bhattacharya and Lab members

NASA Ames Research Center

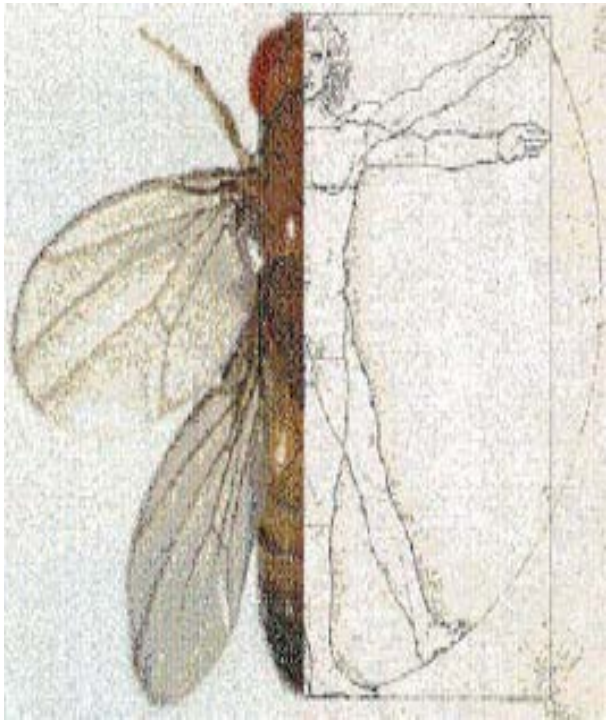


E. Tu, J. Bajpayee et al visit
June 2017

WHY DROSOPHILA ?

- Conserved basic biological processes between organisms.
- Genetically tractable.
- Genetic homology with humans
- Of the known human disease genes , 75% are closely related to *Drosophila* genes

[Reiter et al., 2001, OMIM: Homophila; and Genome Res]



For Spaceflight

- High “n” number
- Genetically identical animals
- Low resource requirements
- Short life cycle - multiple generations
- Measure response of a whole multicellular animal
- Flies used as a model for infection with human pathogens

Hosamani & Bhattacharya 2014

Current Space Flight Projects + Ground Study Projects in Lab

- **Peer-reviewed and competitively selected flight proposals – recent flight missions:**
  - **(a) Heart Flies on SpX-3 (Apr 2014)**: **Space Florida Announced ISS Research Competition Winners at ASGSR, 2012.** Space Florida (the state's aerospace development organization and spaceport authority) and NanoRacks, LLC funded and organized the competition.
    Collaborators: Peter Lee (Stanford U/Ohio State; Bodmer & Ocorr (Sanford Burnham Medical Rsch Institute); Bhattacharya (Ames)
  - **(c) FFL-02 on SpX-11 (Early 2017)**: Space Biology NRA call from 2012 (NNH12ZTT001N): The Effects of Microgravity on Cardiac Function, Structure and Gene Expression using the Drosophila Model. (Bodmer, Ocorr, Bhattacharya, Lee)
  - **(d) FFL-03 on SpX-13 (2017):** Space Biology NRA call from 2014 (NNH14ZTT001N): Does spaceflight alter the virulence of a natural parasite of Drosophila? (Govind, Bhattacharya, Kawaguchi)
  - **(e) Biosentinel on EM1 mission (2018):** High earth orbit/deep space radiation effects on DNA double strand breaks **(Col's Tore Straume and Terry Lusby)**
- **Tech demo or validation missions:**
  - **(f) Fruit Fly Lab hardware (FFL-1) on SpX-5 (Jan 2015)**: Validation of FFL hardware with centrifuge, camera, circadian light, multigenerational growth. Change in virulence of bacterial pathogen
  - **(b) Ames student Fruitfly Experiment AFEx on SpX-4 (Sept 2014) and SpX-9 (2016/2017)**:
    Neurobehavioral experiment on SpX-4 follow up on **SpX-9 (2016?) [Nanoracks & ASGSR]**
- **Hypergravity ground studies & spaceflight studies:**
  - **(g) Changes in immune function in fly host and virulence of bacterial pathogen**
  - **(h) Oxidative stress genes in the fly brain** and effects on behavior under altered gravity conditions

(*Will present more details on topics marked in green text today)

HEART FLIES experiment flew on SpX-3 mission (April – May 2014)

<u>Collaboration:</u>

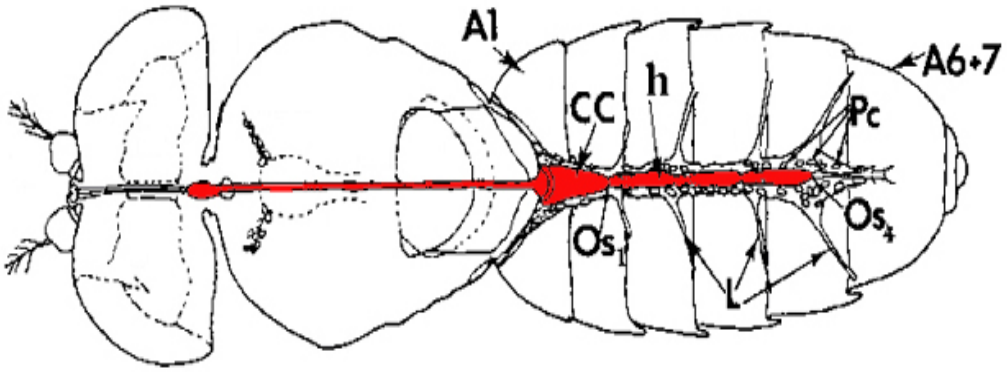
Karen Ocorr – Sanford Burnham Medic Research Instit

Rolf Bodmer – " " " "

Peter Lee – Stanford Univ/Ohio State U

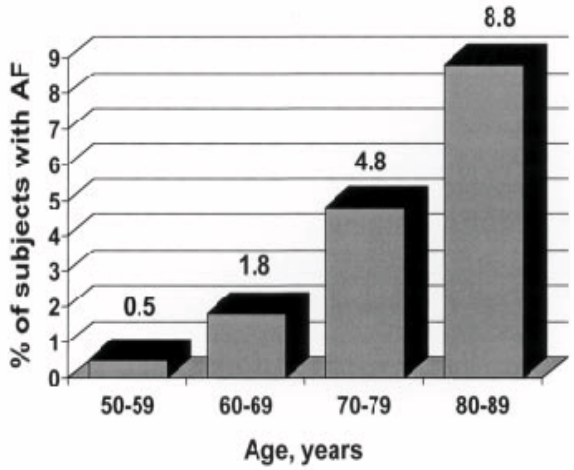
Sharmila Bhattacharya – NASA Ames Research Ctr

The Drosophila Heart Model: an example of the value of the fruit fly for spaceflight studies



Fly hearts show AGE-related arrhythmias

Prevalence of Human Atrial Fib in Framingham Study



Wolf et. al., 1991

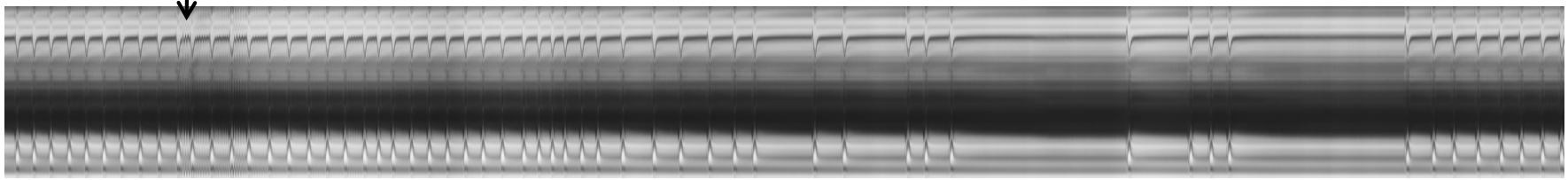
1 week fly (~10 years old)

7 week old fly (~70 years old)

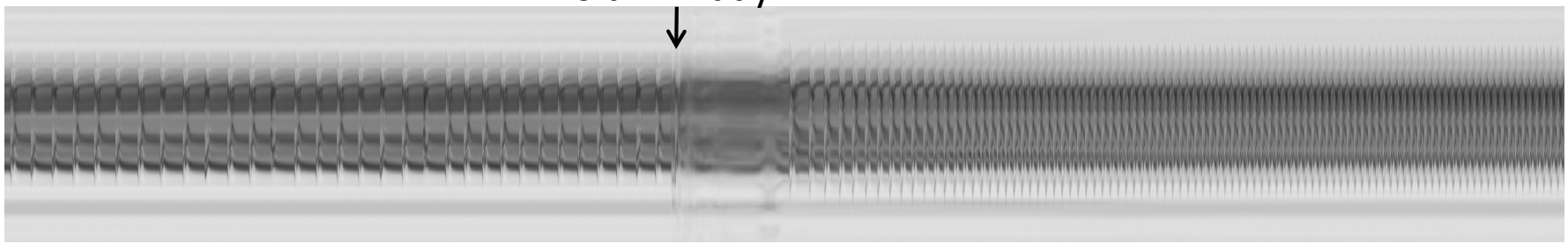
Courtesy K.Ocorr & R.Bodmer

Responses to Applied Drugs/Hormones: Like Humans!

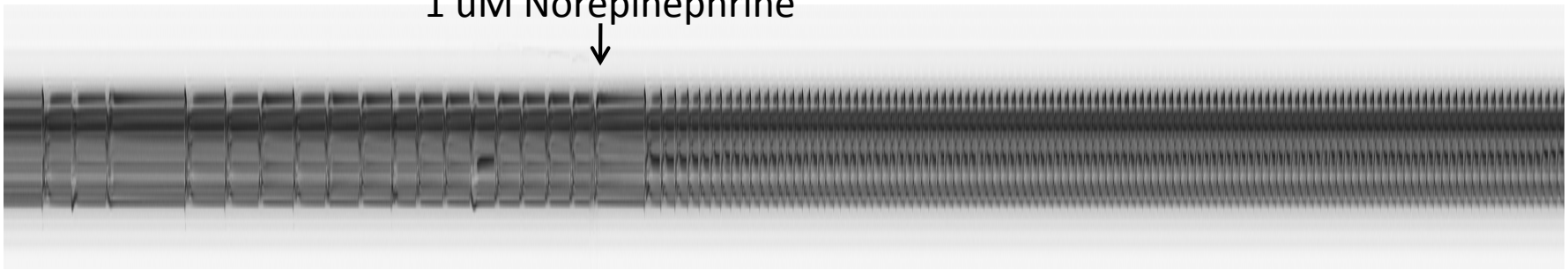
1 uM Clofilium (Cardiac Drug to treat Tachycardia)



10 uM Bradykinin



1 uM Norepinephrine



Courtesy K. Ocorr & R. Bodmer

Drosophila Cardiac Model flown on FFL – 02/SpX-11 (June to July 2017)

Five *Drosophila melanogaster* genotypes to be tested:

1. CantonS (wildtype)

2. w^{1118} (wildtype)

- Flies have similar genes/proteins as human hearts (we share 75% of disease causing genes)
- Flies show cardiac aging effects similar to humans.
- Each line has a different genetic background but All Flies from a single line are the same genetically.
- Thus, common effects seen in both lines must be due to μG not genetic background.

3. *KCNQ*^{186} (ion channel mutant)

- Exhibits arrhythmias similar to human Long QT Syndrome.
- Athletes with un-diagnosed mutations in this gene (and other related channels) often die of sudden cardiac arrest under exertion.

4. D45 (myosin mutant)

- Mutations in this gene contribute to hypertrophic cardiomyopathies in both flies and humans.
- Hearts from this "slow" myosin mutant are dilated but also show reduced cardiac aging.

5. PQ46 (GFP-tagged PolyQ)

- μG disrupts Protein Homeostasis in hearts from both wildtype lines (1&2).
- We can monitor protein misfolding directly in this line.

FFL-02 in the Press

The New York Times https://nyti.ms/2rzuv5q

SCIENCE

Fruit Flies and Mice to Get New Home on Space Station, at Least Temporarily

By KENNETH CHANG JUNE 2, 2017

A bit of science trivia: Did you know that the heart of a fruit fly beats at about the same pace as yours?

That's among the reasons that 400 adult fruit flies and 2,000 eggs are packed to go to the International Space Station, for an experiment on long-term weightlessness and how it might affect the cardiovascular health of astronauts.

La Jolla Lab to Send Fruit Flies into Space

By NBC 7 Staff

San Diego Researchers Will Send Fruit Flies Into Space for Science

Researchers at Sanford Burnham Prebys Medical Discovery Institute will send boxes of fruit flies (Drosophila melanogaster) to the International Space Station today to study the impact of weightlessness on the heart. The fruit flies are

Fruit flies flying to space to help scientists study the human heart

Falcon 9 rocket to launch three tons of supplies, experiments Saturday

By James Sparvero [https://www.clickorlando.com/author/james.sparvero] Reporter

Posted: 6:58 PM, June 02, 2017
Updated: 6:34 PM, June 02, 2017

Join the conversation

TITUSVILLE, Fla. - About 6,000 pounds of supplies and science experiments scheduled for launch to the International Space Station [http://www.clickorlando.com/topic/International Space Station#entityid=4205051404&lang=en] Saturday will include what you might normally consider nothing more than a nuisance.

But Dr. Karen Ocorr, a scientist studying heart disease, told News 6 studying the effects of zero gravity on a fruit fly's heart can reveal a lot about the human heart as well.

Fruit Flies to Be Launched to International Space Station

NASA Ames

June 1, 2017

Fruit Fly Lab (FFL-02) Scientist's Blog

Christina Cheung is one of the support scientists for NASA's Fruit Fly Lab and the outreach lead for the Space Biosciences Division at NASA's Ames Research Center in Silicon Valley. Christina is blogging from Ames and from Kennedy Space Center in Florida about her experiences during the pre-flight, flight, and post-flight periods of the FFL-02 experiment. It is scheduled to launch to the International Space Station in June 2017 aboard SpaceX's eleventh commercial resupply services mission.

Post 0 • Post 1 • Post 2 • Post 3 • Post 4

Post 5 • Post 6 • Post 7 • Post 8 • Post 9

Post 10 • Post 11 • Post 12 • Post 13 • Post 14

Post 15 • Post 16

NASA Ames @NASAAmes

Before sending fruit flies to the @Space_Station, we first have to separate the males from the females. Learn more: go.nasa.gov/2pUwem4

6:07 PM - May 26, 2017

JUNE 15, 2017

SPACEFLIGHT NOW

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BREAKING NEWS > [June 15, 2017] Live coverage: Previously-flown Falcon 9 rocket fires engines on Florida

SpaceX rocket again set for station delivery after scientists swap mice, fruit flies

June 3, 2017 Stephen Clark

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REPORTAJE

Moscas, gusanos y peces. Nuestros 'dobles' de laboratorio

Más allá de ratones o chimpancés, numerosos estudios con otros animales están consig... enfermedades como el cáncer o la regeneración cardiaca. Las moscas de la fruta, ¡ca... comparten con nosotros mucho más de lo que imaginas.

SBP Sanford Burnham Prebys MEDICAL DISCOVERY INSTITUTE

Fruit Flies Journey to International Space Station to Study Effects of Zero Gravity on the Heart

Study led by SBP researchers could help uncover therapeutics to prevent or treat heart conditions, both in space and on Earth

Why Do Scientists Use Fruit Flies in Research?

Seiworthy

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Published on May 17, 2017

BMSIS Young Scientists Christina Cheung and Imar Hamid talk about fruit flies. Why do scientists use them for experiments and how do they teach us about humans?

How Fruit Flies Help us Understand Human Responses to Microgravity

NASA Johnson

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ISS Research @ISS_Research

1,000 bottles of fruit flies are packed & ready to head to @Space_Station for cardiovascular @ISS_Research. go.nasa.gov/2pUwem4

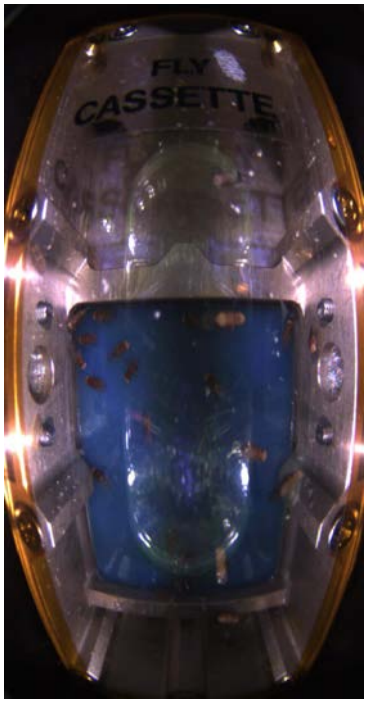
NASA Ames and 4 others

10:11 AM - May 22, 2017

Spaceflight causes increased virulence of *Serratia marcescens* (Db11) on a *Drosophila melanogaster* host

(Results from our FFL-01 mission on SpaceX – CRS 5 mission, launched Jan 10, 2015)

FLIES ARE AN IMPORTANT IMMUNE MODEL SYSTEM FOR SPACEFLIGHT STUDIES



- ***Drosophila melanogaster* (fruit fly)** is an excellent model of the innate immune system and displays decrements in immune function following spaceflight, similar to that seen in humans post-flight

  (See our paper *Marcu et al, 2011* from the shuttle flight STS-121).
- Alterations in bacterial virulence and antibiotic resistance during spaceflight has long been a point of concern for NASA.

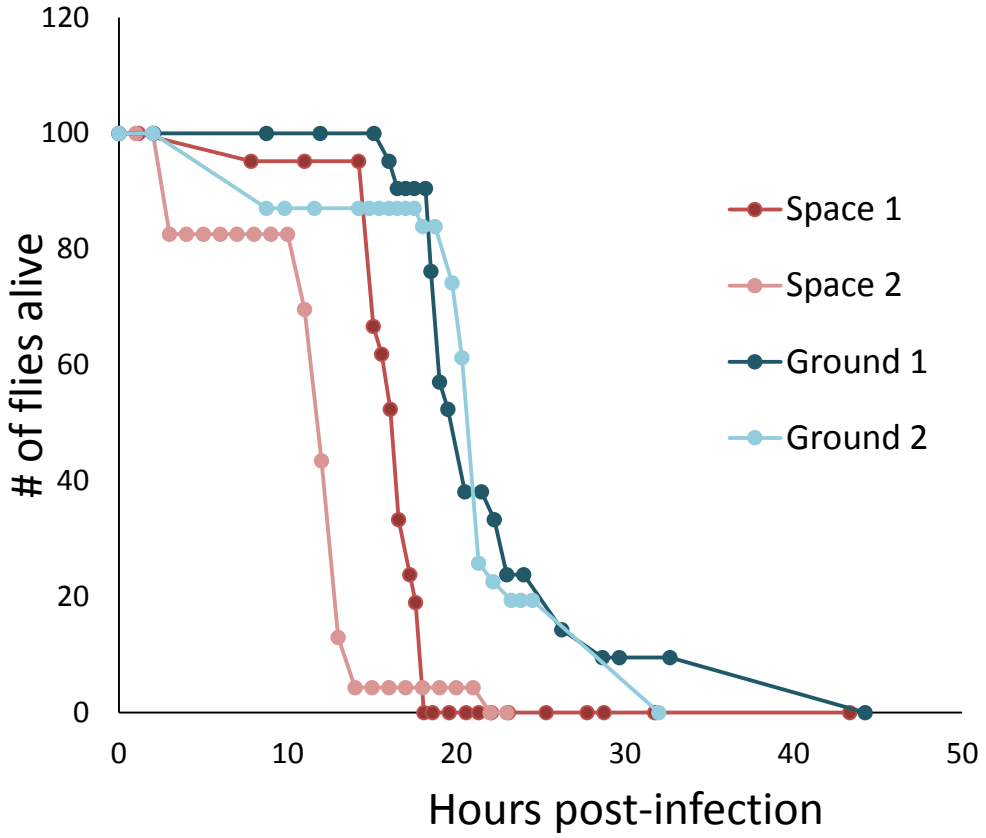
  *(*See *Wilson et al, 2007 and 2008; Crabbe et al, 2011; etc*)
- ***Serratia marcescens*** is a gram-negative opportunistic pathogen to humans typically causing nosocomial infections.
- The *S. marcescens* sub-strain Db11 is a pathogen for *Drosophila melanogaster*.
- *S.marcescens* has been found in free floating water condensates on ISS

  (See *Ott et al, 2004*)
- We recently flew a sample of *S. marcescens* Db11 as part of separate study and were interested to see if any of the returned bacterial samples displayed altered virulence in *Drosophila* hosts injected post-flight.

Serratia marcescens bacteria
(Castelli ME. et.al. J.Bac-2008)

w^{118} *S. marcescens* Db11 injections

Spaceflight samples overall are 4-6 times more lethal than Ground controls



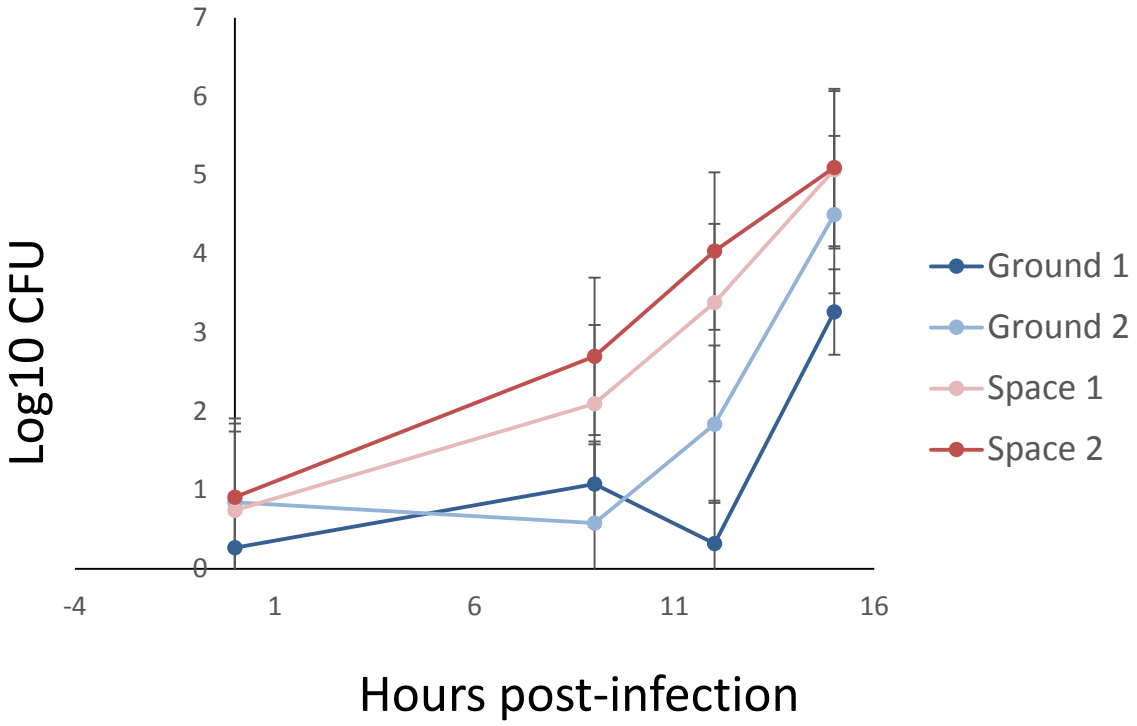
Cox Proportional Hazards test:

Risk Ratios for Treatment

Level1	/Level2	Risk Ratio	Prob>Chisq
Ground 2	Ground 1	1.3477574	0.3652
Space 1	Ground 1	5.526364	<.0001*
Space 1	Ground 2	4.1004144	0.0002*
Space 2	Ground 1	6.4117731	<.0001*
Space 2	Ground 2	4.7573643	<.0001*
Space 2	Space 1	1.1602155	0.6385
Ground 1	Ground 2	0.7419733	0.3652
Ground 1	Space 1	0.1809508	<.0001*
Ground 2	Space 1	0.2438778	0.0002*
Ground 1	Space 2	0.1559631	<.0001*
Ground 2	Space 2	0.2102004	<.0001*

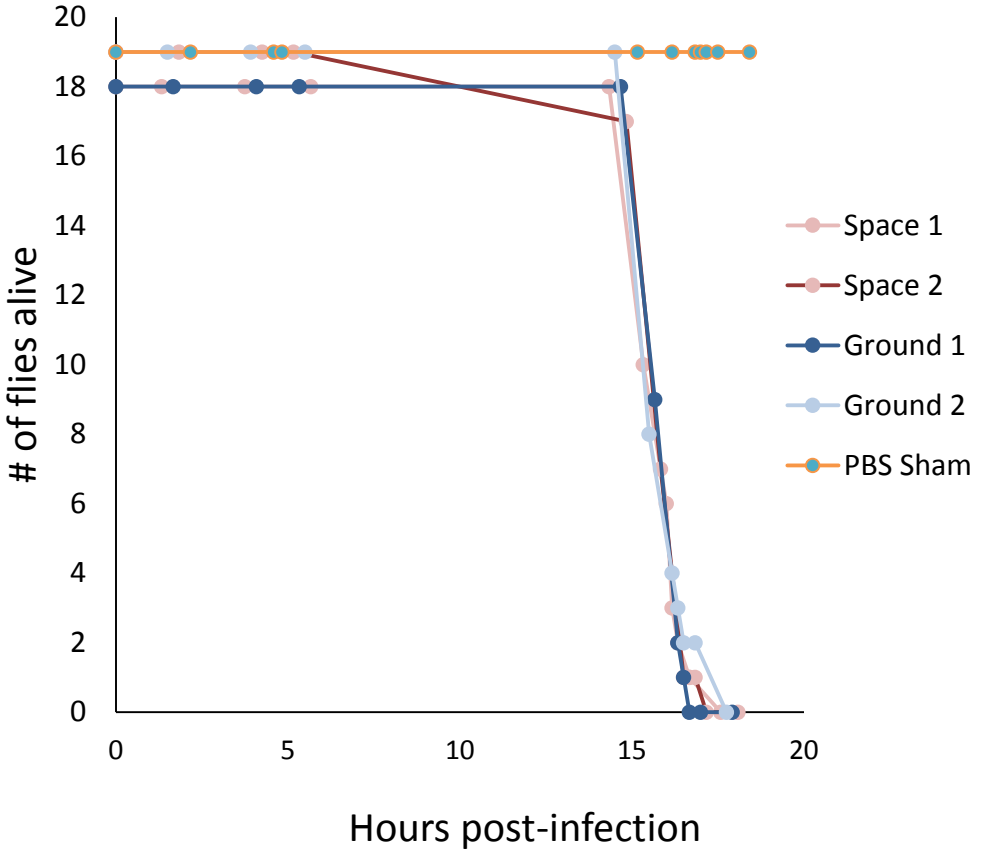
In-host growth of Db11 after injection

There is no significant difference of in-host growth rate between Space and Ground bacteria after infection (F=2.629, P=0.122)



w^{118} survival – 1st subculture with Db11

After culturing spaceflight samples on the ground, they became less virulent, and statistically had the same lethality as Ground controls



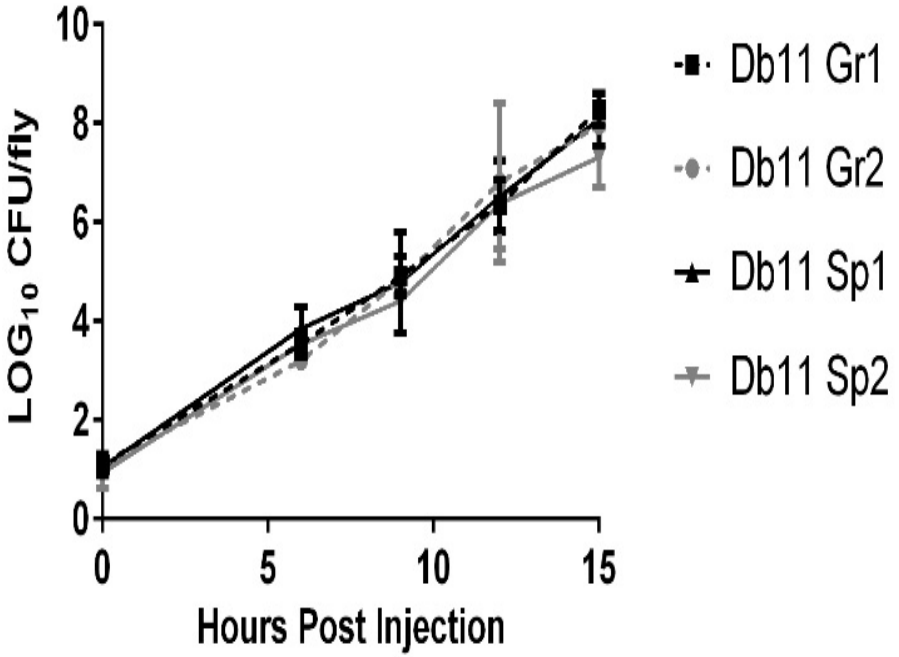
Cox Proportional Hazards test:

Risk Ratios for Treatment

Level1	/Level2	Risk Ratio	Prob>Chisq
Ground 2	Ground 1	0.4217154	0.0883
Space 1	Ground 1	0.9674476	0.9387
Space 1	Ground 2	2.2940772	0.0991
Space 2	Ground 1	0.8860095	0.7939
Space 2	Ground 2	2.1009656	0.1068
Space 2	Space 1	0.9158217	0.8478
Ground 1	Ground 2	2.3712676	0.0883
Ground 1	Space 1	1.0336477	0.9387
Ground 2	Space 1	0.4359051	0.0991
Ground 1	Space 2	1.1286561	0.7939
Ground 2	Space 2	0.4759716	0.1068
Space 1	Space 2	1.0919156	0.8478

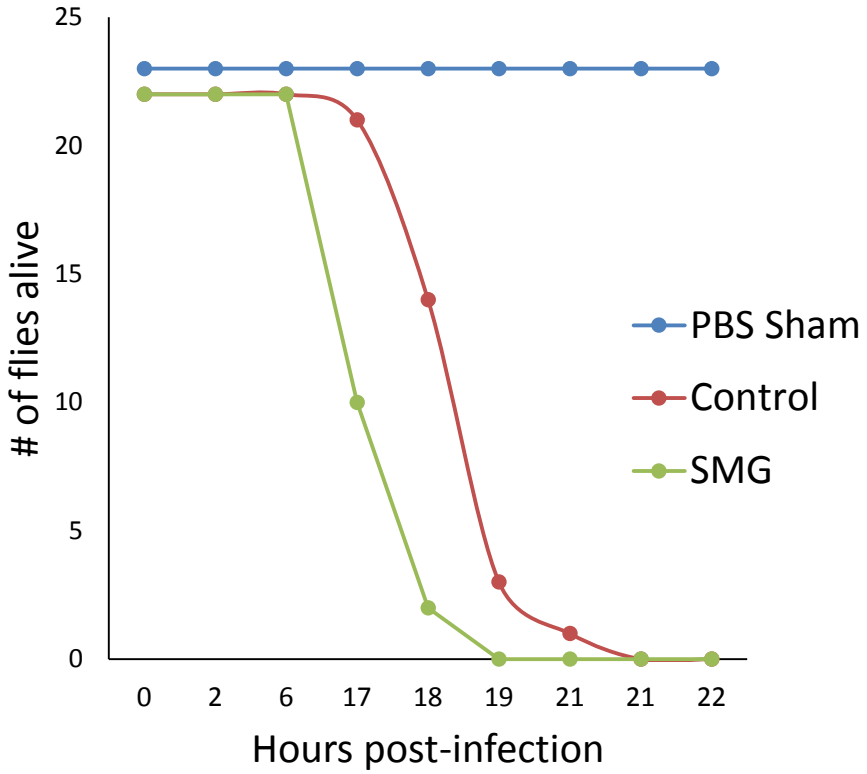
In-host growth of 1st subculture Db11

Again, no significant difference of in-host growth rate between Space and Ground bacteria after infection (F=2.129, P=0.152)



w^{118} survival – simulated microgravity Db11

When grown in low-shear simulated microgravity (rotating wall vessel), there is no significant difference in survival or in-host growth between control and simulated microgravity *S. marcescens*



Cox Proportional Hazards test:

Risk Ratios for Treatment

Level1	/Level2	Risk Ratio	Prob>Chisq
PBS Sham	Control	3.134e-12	<.0001*
SMG	Control	1.4830863	0.4401
SMG	PBS Sham	4.732e+11	<.0001*
Control	PBS Sham	3.191e+11	<.0001*
Control	SMG	0.6742696	0.4401
PBS Sham	SMG	2.113e-12	<.0001*

Host response to infections with Db11

Genetic (RNA gene expression) comparison of flies infected with Space versus Ground strains of *S. marcescens*

All values P<0.05

Gene name	Fold change	Description
Peptidoglycan recognition protein LB	1.42	Gram-negative bacteria defense
vsg (visgun)	5.48	Broadly involved in cell proliferation
Acid phosphatase 1	5.04	Clinical role in human disease, precise function unknown
Longitudinals lacking (lola)	2.96	Involved in axon guidance
Methyltransferase	2.41	Broadly involved in disease and metabolic disorders
Lectin-like gene	2.00	Pattern recognition receptor, immune response
Trypsin (serine protease)	1.02	Melanization response to pathogens/parasites

Conclusions

- A space flown sample of *Serratia marcescens* Db11 displayed increased virulence compared to ground controls when injected into adult female fly hosts post-flight.
- This is a similar result to Nickerson et al's post-flight data with *Salmonella* infections of mice
- Simulated microgravity treatment of Db11 (rotating wall vessel grown cells) do not show the same increase in virulence as spaceflight treated samples
- Molecular biological analyses (e.g. RNAseq of spaceflown and ground control Db11 infected flies) shows an alteration in host immune genes infected with spaceflight and ground bacterial samples
- We have previously shown that the immune system show decrements in the fly host after spaceflight, we now show that *Serratia marcescens* Db11 displays increased virulence after spaceflight when infecting fly hosts postflight. The fruit fly remains an important translational model organism in studying spaceflight effects both on host immune function and for testing virulence of pathogens

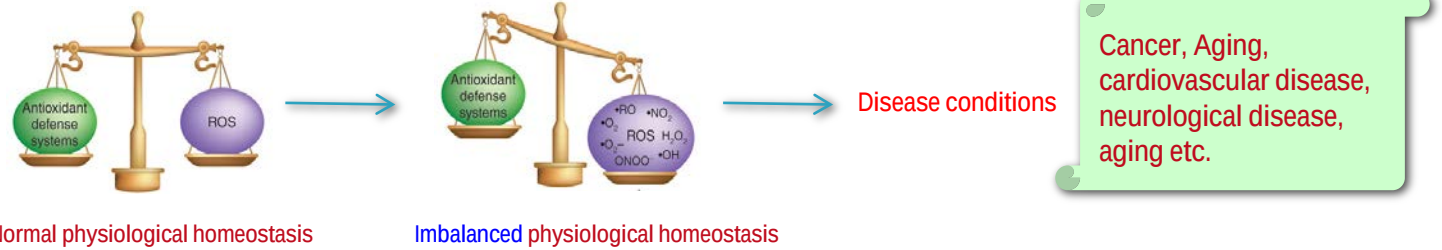
Altered Gravity Induces Oxidative Stress in *Drosophila melanogaster.*

Sharmila Bhattacharya
Ravikumar Hosamani
Anastasia Vavilina
Maximilien Baas-Thomas
Iman Hamid
Andrew Pelos

NASA Ames Research Center, CA

OXIDATIVE STRESS PATHWAY IN *Drosophila melanogaster* (OR FRUIT FLY) SIMILAR TO MAMMALIAN AND OTHER MODELS

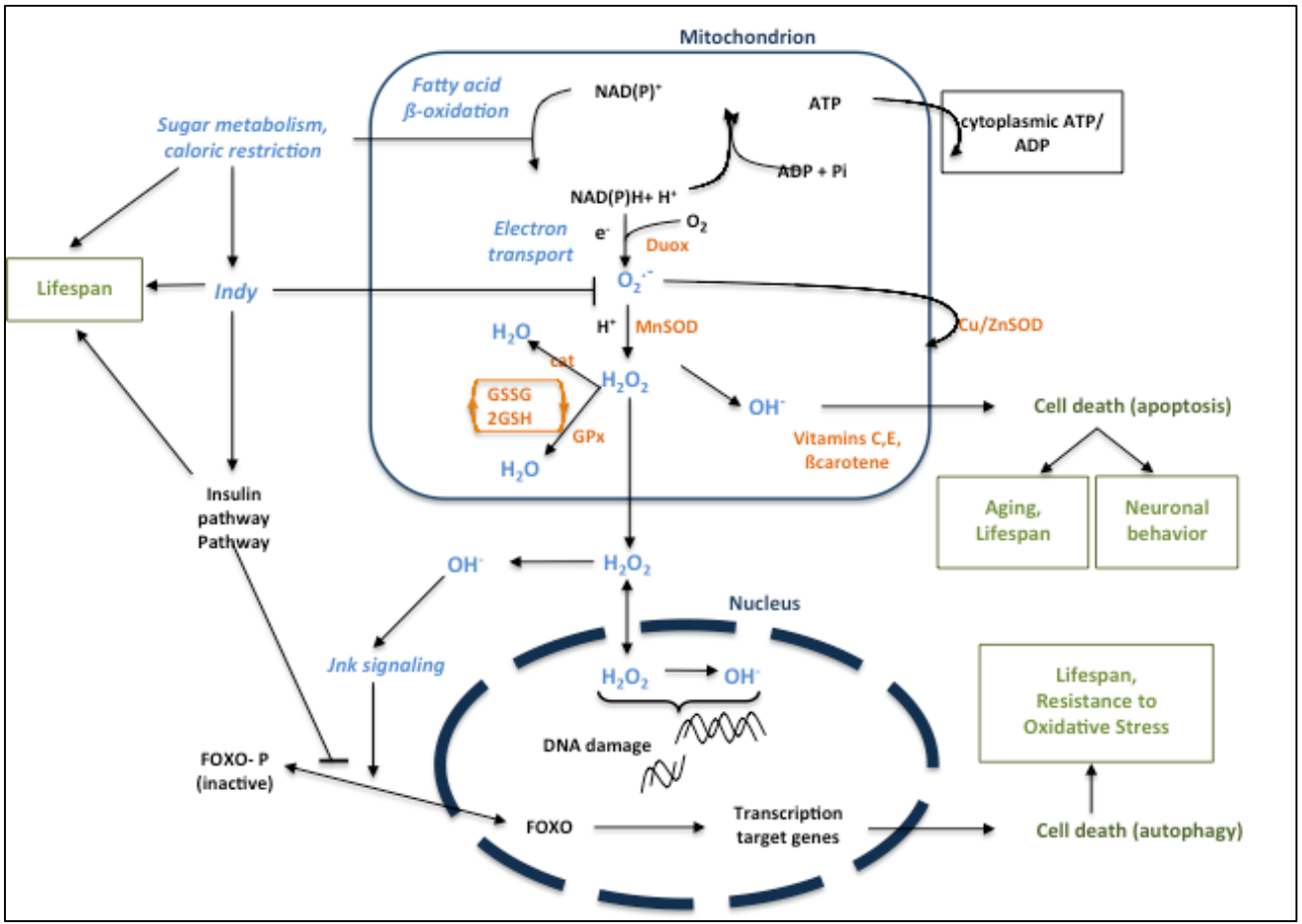
❖ OXIDATIVE STRESS



Normal physiological homeostasis

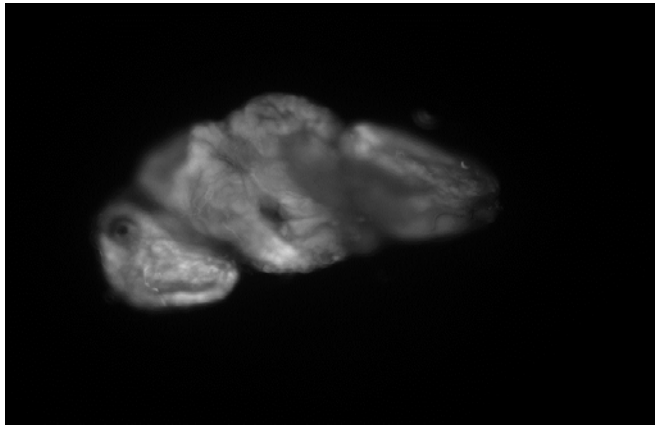
Imbalanced physiological homeostasis

- Evolutionarily basic physiological oxidative stress pathway conserved across species
- Main difference in glutathione-dependent antioxidant defense in *Drosophila* and mammalian models: glutathione reductase is replaced by the thiol redox enzyme system in *Drosophila*

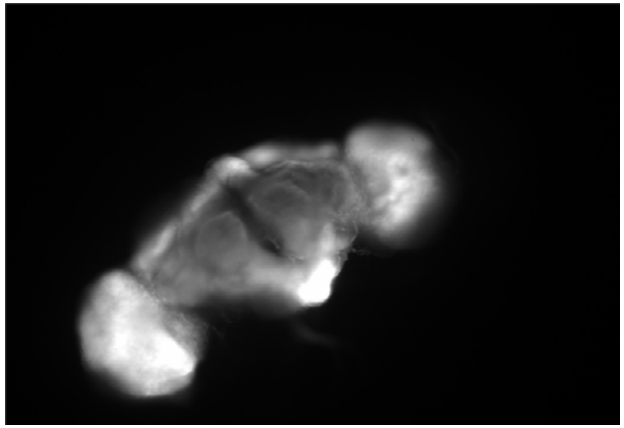


HYPERGRAVITY EXPOSURE, ROS PRODUCTION, AND LOSS OF DOPAMINERGIC NEURONS IN FEMALE ADULT FLY HEADS

ROS PRODUCTION IN THE BRAIN



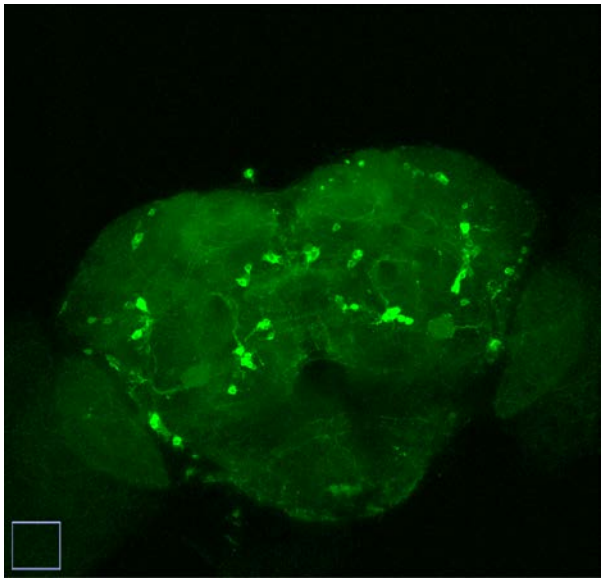
Normal ROS (1g)



Elevated ROS (3G)

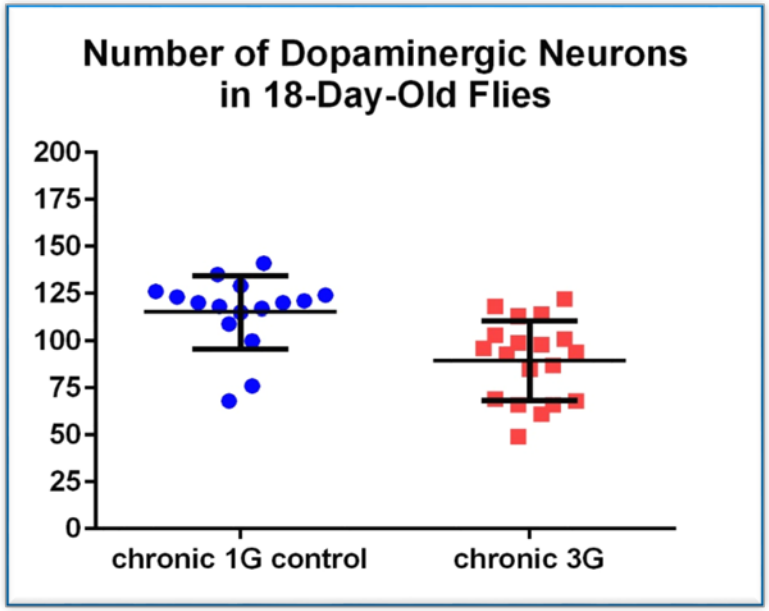
ROS experiments showing normal vs. elevated ROS levels, as indicated by increased fluorescence using DCFH-DA . (Images taken with Zeiss fluorescence microscope at 10x magnification)

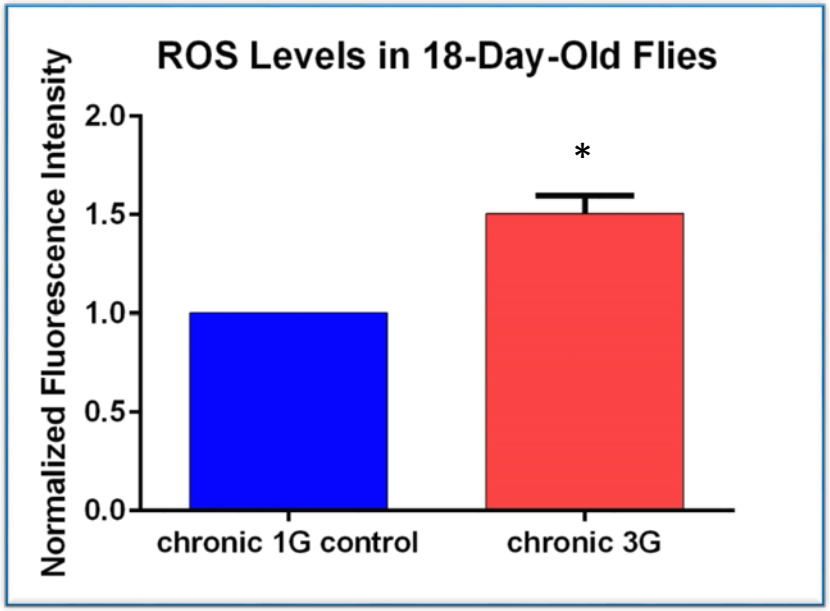
DOPAMINERGIC NEURONS VISUALIZED IN THE FLY BRAIN WITH GFP



(Image taken with a Leica SP5 confocal microscope at 20x magnification.)

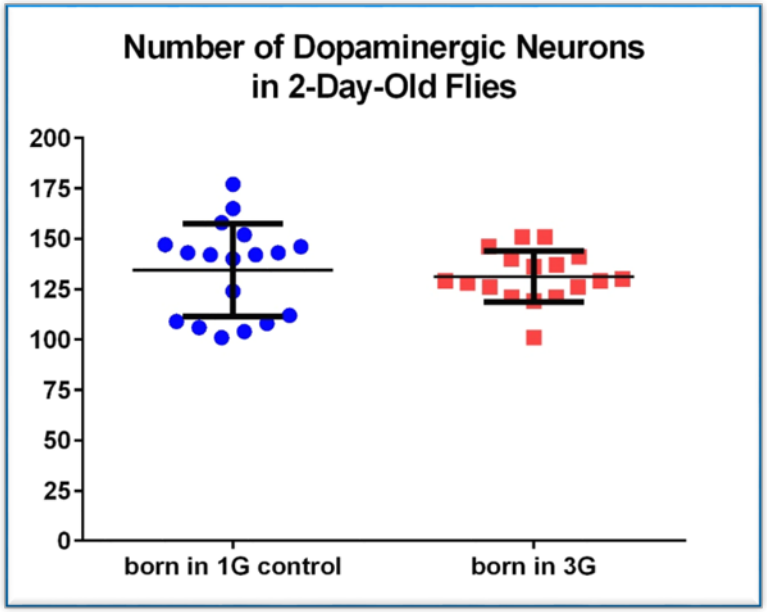
DECREASE OF DOPAMINERGIC NEURONS & INCREASE IN ROS LEVELS IN FLY HEADS FROM 18 DAY OLD FEMALE FLIES EXPOSED TO 3G HYPERGRAVITY AS ADULTS

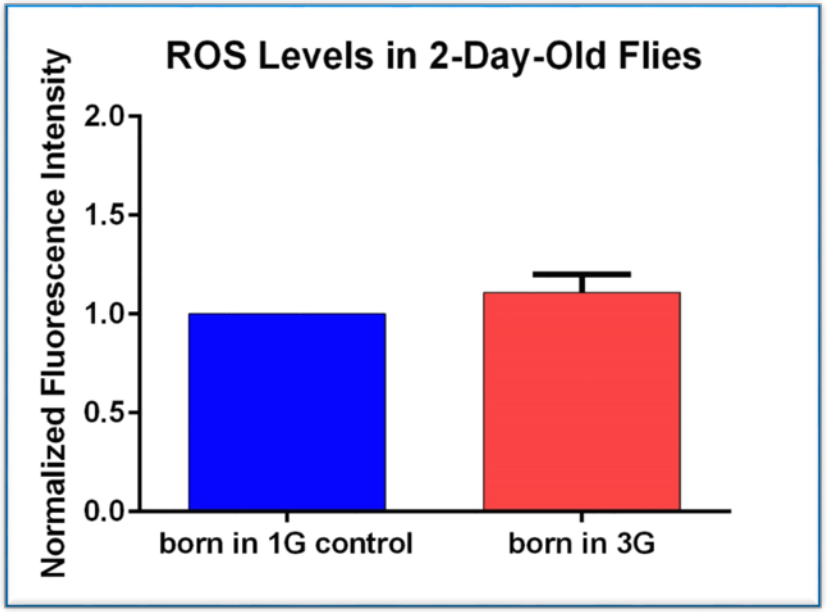




The group exposed to chronic 3G shows a significant (Mann-Whitney, p=0.01) increase in ROS levels in the brain, correlated with a significant (Mann-Whitney, p=0.002) decrease in the number of dopaminergic neurons in the brain.

NO CHANGE IN DOPAMINERGIC NEURONS NOR IN ROS LEVELS IN FLY HEADS FROM 2 DAY OLD FEMALE FLIES EXPOSED TO 3G HYPERGRAVITY FROM EMBRYONIC STAGE

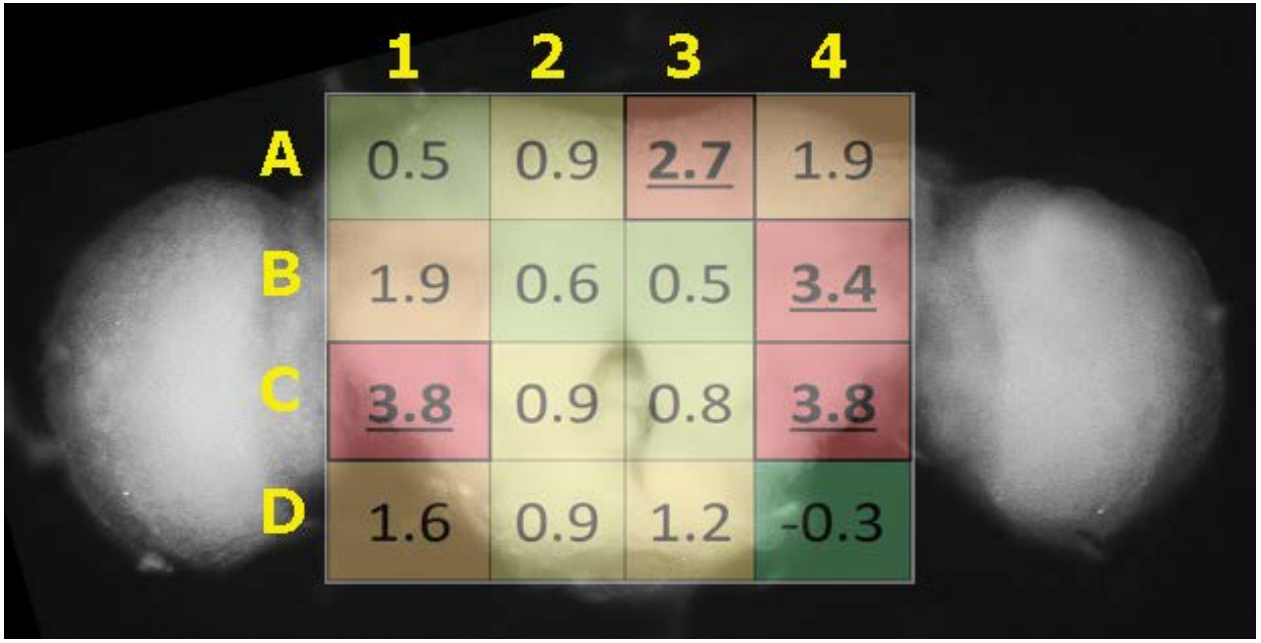




Flies born from eggs laid in 3G show no significant alteration in the number of dopaminergic neurons or ROS levels in the brain

WORK AHEAD: Will test 18 day old adults exposed to hypergravity throughout development

LOCALIZATION OF THE SPECIFIC AREAS IN THE BRAIN FROM WHERE DOPAMINERGIC NEURONS ARE LOST IN HYPERGRAVITY ARE SIMILAR TO REGIONS AFFECTED BY OXIDATIVE STRESS

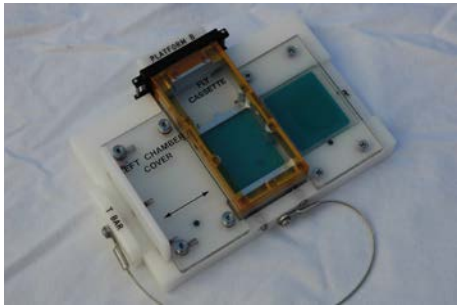


Values show number of neurons lost per region, with underlined values showing areas of significant change ($p<0.005$).

Conclusions for Oxidative Stress Studies:

- **Good translational model:** *Drosophila* oxidative stress pathway is conserved with mammalian and other vertebrate systems in terms of overall pathway and function
- Both spaceflight **and hypergravity induces oxidative stress** in *Drosophila melanogaster*
- Hypergravity treatment causes increased **ROS production in the brain and increased loss of dopaminergic neurons** similar to that seen in other studies where only an increase in oxidative stress can cause similar loss of dopaminergic neurons. (Good translational value – flies used as Parkinson's Disease model for studies of oxidative stress and effect of pesticides like rotenone on DA neuron loss)
- **Developmental stage at time of hypergravity exposure may matter**: The above effect is seen in adults exposed to hypergravity, but not in young animals raised under hypergravity conditions throughout development

A few examples of past spaceflight studies with fruit flies from NASA Ames



- "FIT" expt flew on STS-121 (shuttle mission in 2006, & on SpaceX-CRS 5 in 2015)
- Showed that spaceflight affected innate immune function in flies, similar to humans
- Used flies to understand underlying molecular mechanism of immune changes in space
- Published in *PLoS ONE 6(1): e15361. 2011. doi:10.1371/journal.pone.0015361*

- HEART-FLIES flew on SpaceX-CRS 3 (2014)
- Used flies to study cardiovascular changes resulting from spaceflight
- Hardware built entirely by students
- Partnered with Sanford Burnham Prebys Medical Discovery Institute (San Diego); Space Florida Grant Consortium; Nanoracks LLC; Stanford University; Ohio State University

- AFEx flew on SpaceX-CRS 4 (2014)
- Studied behavioral changes in flies in space
- Hardware built by students
- Partnered with ASGSR (Am Soc for Grav & Space Res); STC (Science & Tech Corp.) & Nanoracks LLC to launch experiment

Benefit to the Larger Community

- All fruit fly hardware and experiments flown by NASA between 1999 and 2017 have been developed either completely by or in collaboration with NASA Ames Research Center. **Ames is the only NASA center with expertise in spaceflight research using fruit flies**

- **More than 61 students trained** (just from fruit fly experiments) in the last 18 years and they participated in aspects of development, integration, spaceflight and post-flight data analysis of the spaceflight experiments

- **Over 120 recent news/media/web articles** relating to fruit fly research in space! The following **Youtube video alone has been viewed 63,451 times!**
  https://www.youtube.com/watch?v=ArHDSjfKDAs

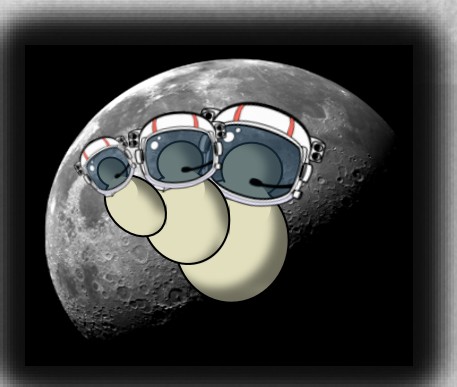
- Upcoming experiments : Fruit Fly Lab-02 (FFL-02 on SpaceX-CRS 11 in 2017); and FFL-03 (2018)

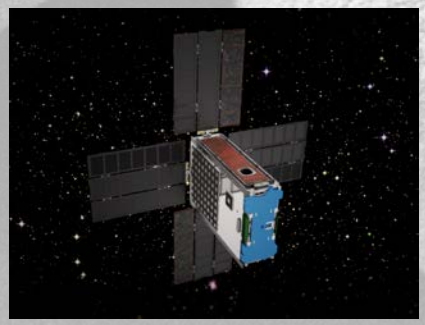
- **NASA Ames collaborate(d)/partner(ed) with**:
  - **Universities:** e.g. Yale University (NIH-B1 on STS-93); UC Davis ("FIT" on STS121); Stanford University (HEART-FLIES on SpX-CRS 3); City University of New York (FFL-03 on SpX-CRS 11); Ohio State University (HEART FLIES and FFL-02); Rice University (EMCS fruit fly) etc
  - **Research and other Institutions:** e.g. Sanford Burnham Prebys Medical Discovery Institute (HEART-FLIES on SpX-CRS 3, and FFL-02); Florida Space Consortium (HEART-FLIES); etc
  - **Commercial entities/companies**: e.g. Bioserve (NIH-B1, CSI spider habitat, FFL-02 and FFL-03); Nanoracks LLC (FFL-01 on SpX-CRS 5, HEART-FLIES on SpX-CRS 3, AFEx on SpX-CRS 4); Science and Technology Corp (AFEx on SpX-CRS 4), Space X (HEART-FLIES, AFEx, FFL-01, FFL-02, FFL-03); Techshot (Multivariable gravity platform hardware from 2017 onwards); Airbus (FFL-01); Intrinsyx (FFL-01); Wyle Inc (FFL-01, FFL-02, FFL-03); etc

Using brewer's yeast as a "canary in the coal mine" to prepare for long term human exploration of deep space

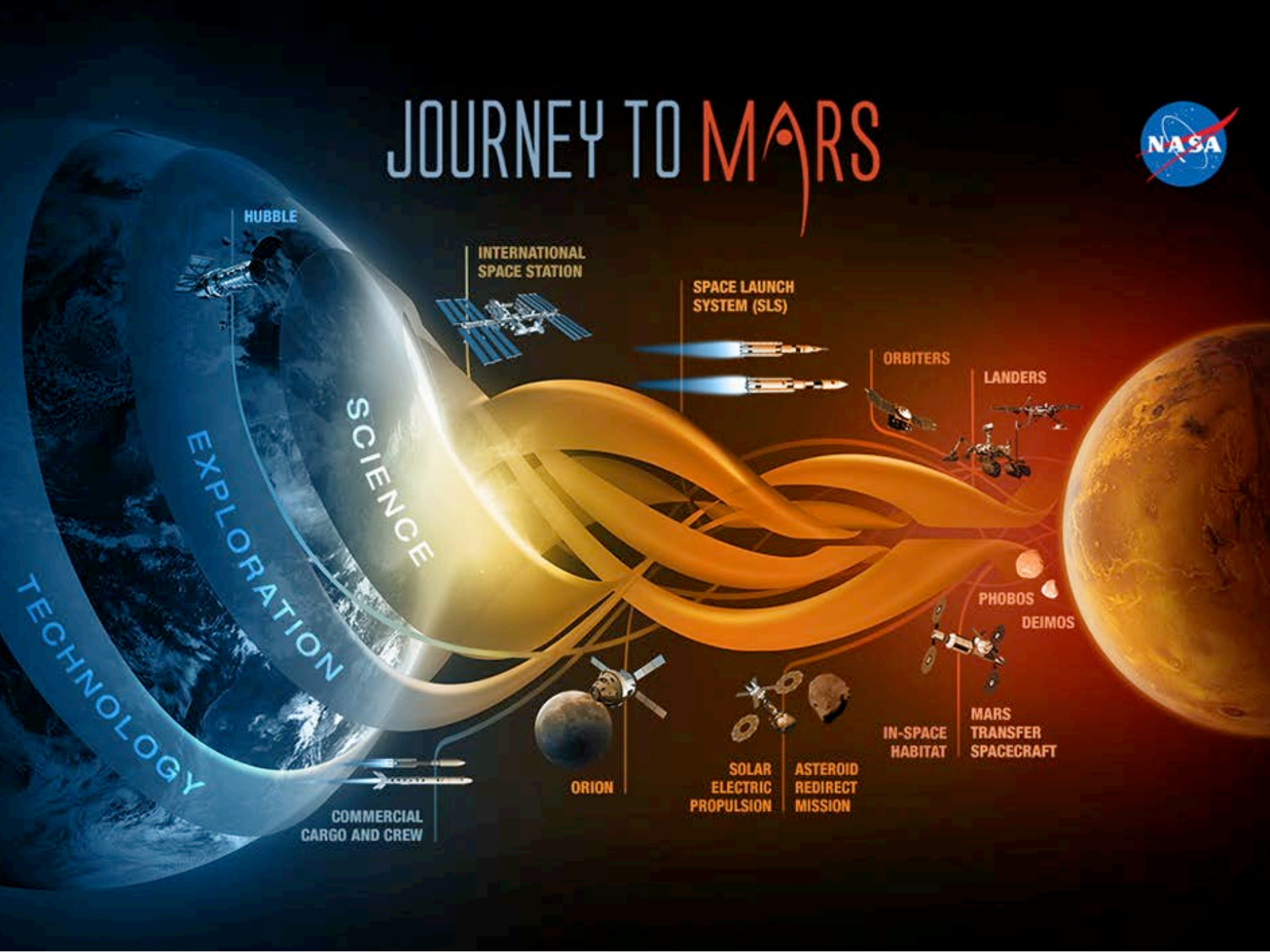
Sharmila Bhattacharya

NASA Ames Research Center





Interplanetary Small Satellite Conference. San Jose, CA, May 1-2, 2017


JOURNEY TO MARS
NASA
HUBBLE
INTERNATIONAL SPACE STATION
SPACE LAUNCH SYSTEM (SLS)
ORBITERS
LANDERS
SCIENCE
EXPLORATION
TECHNOLOGY
PHOBOS
DEIMOS
IN-SPACE HABITAT
MARS TRANSFER SPACECRAFT
ORION
SOLAR ELECTRIC PROPULSION
ASTEROID REDIRECT MISSION
COMMERCIAL CARGO AND CREW

Radiation & spaceflight damages chromosomes in astronauts' white blood cells (lymphocytes) – expect effect to be greater in deep space than on ISS

F.A. Cucinotta, M.Y.Kim, V. Willingham, and K.George. (2008) Rad Res. 170:127. (Studies in ISS, Mir & STS astronauts)

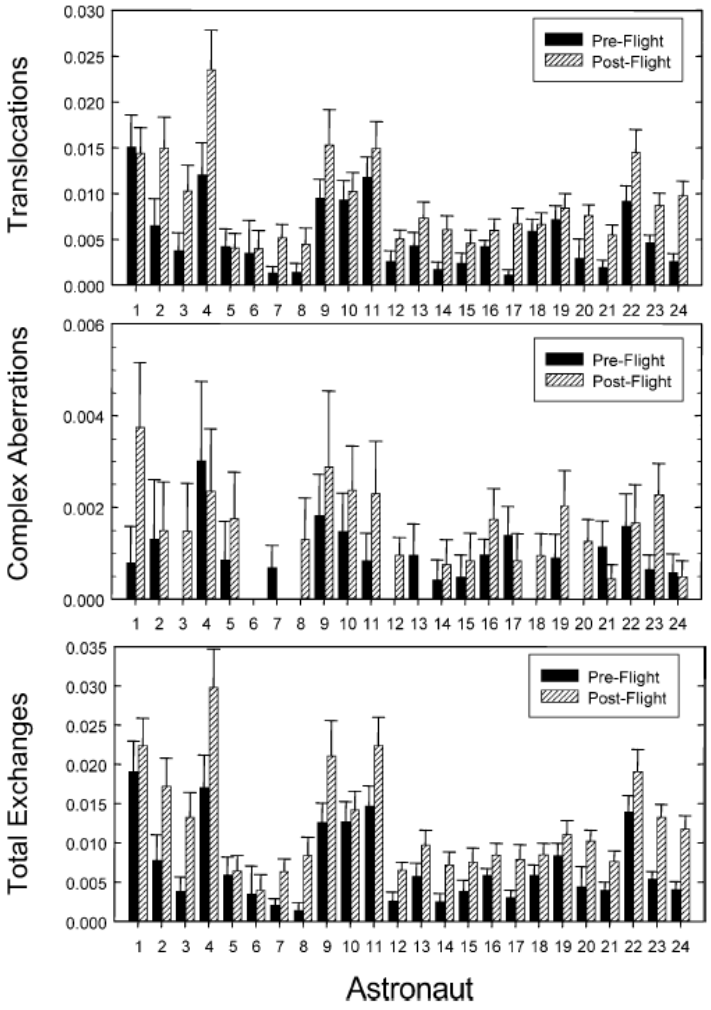
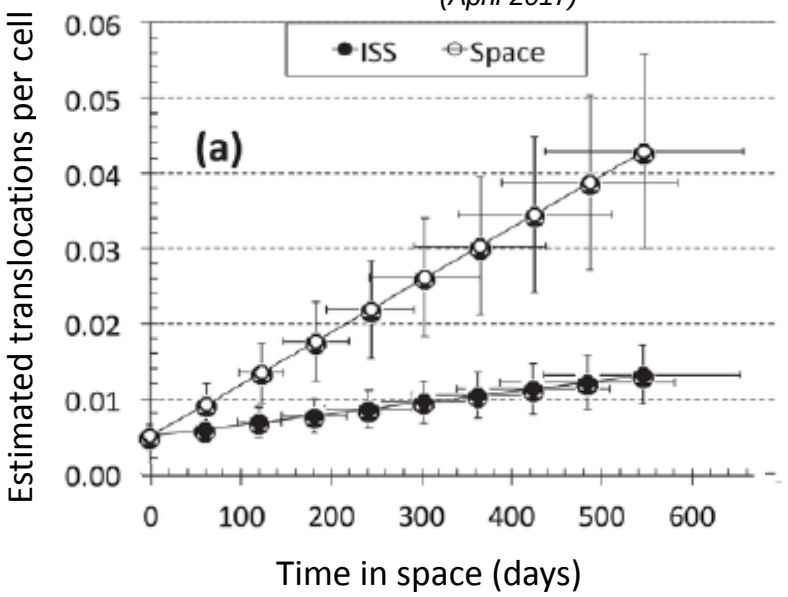


FIG. 4. Frequency of translocations, complex aberrations or total chromosome exchanges measured in each astronaut's blood lymphocytes before and after his or her respective space mission on ISS, Mir or STS. Increases in total exchanges were observed for all astronauts. Translocations (22 of 24) and complex aberrations (17 of 24) were increased in the majority of astronauts.

T.Straume, T.C. Slaba, S. Bhattacharya, L.A. Braby. (2017) Life Sci Space Res. (April 2017)

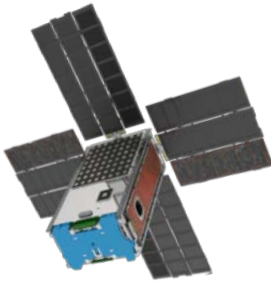


- Astronauts have ↑ in # chromosomal abnormalities, even at low Earth orbits, during ISS, Mir & STS (Hubble shuttle) missions
- The relative increase in frequency of these chromosomal abnormalities ranges from 1.5 to 1.8 times more than pre-flight levels (95% CL)
- >80% of organ dose equivalents on ISS are from galactic cosmic rays (GCR) which are difficult to shield
    - GCR will be much more abundant as astronauts go to higher orbits beyond Earth's protective magnetosphere

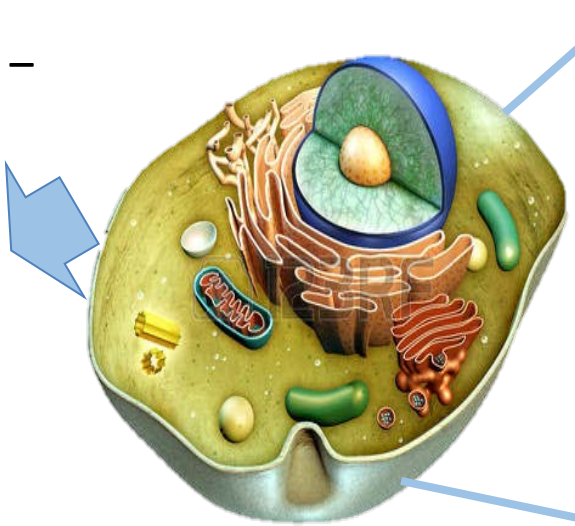
Why do we need a biological radiation sensor in deep space?

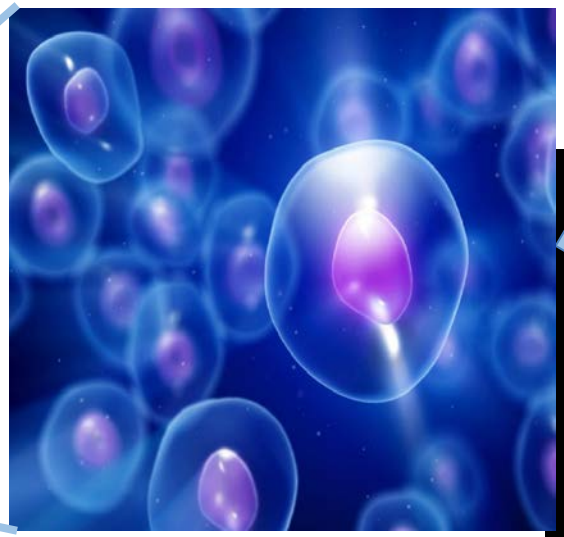
- Ionizing radiation (IR) presents a major challenge to humans and to other biological organisms discussed previously
- Preparation for long-term human exploration and residence in deep space
- Critical knowledge gap: deleterious effects due to long-term exposure to ionizing radiation in space
- Hard to accurately replicate flux and composition of space radiation on Earth
- Microgravity might be a confounding factor, particularly for larger multicellular organisms, when coupled with ionizing radiation in deep space

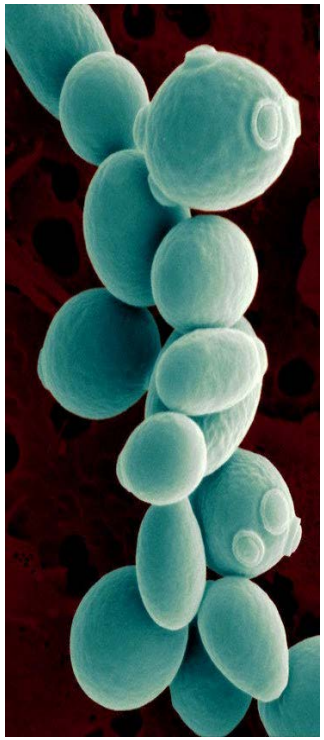
Why Yeast?



Eukaryotic cells – with a "true nucleus"







S. cerevisiae **(budding yeast)**

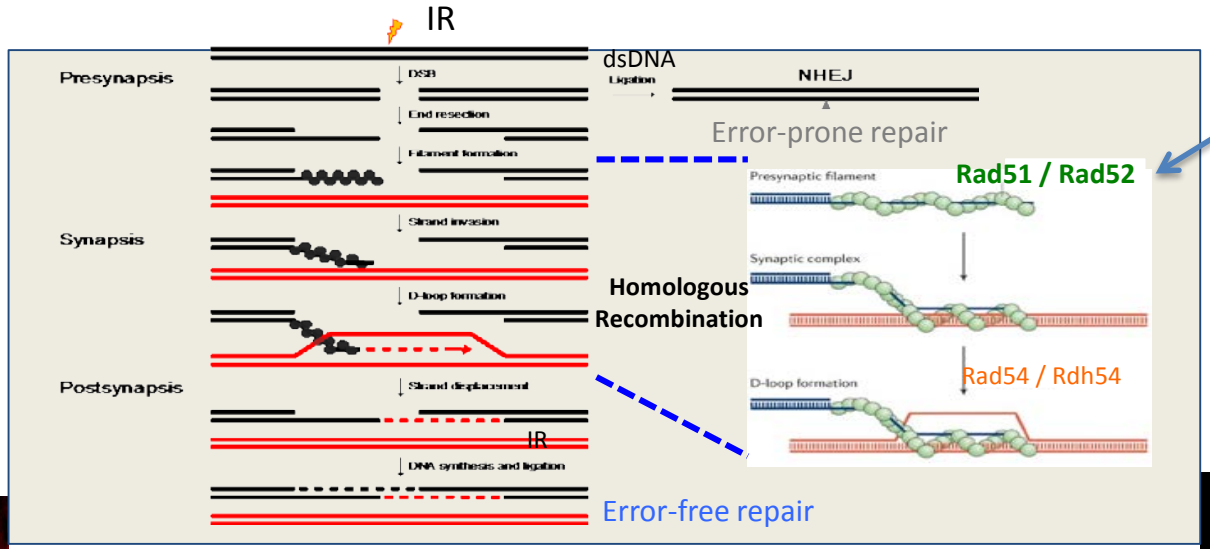
414 essential yeast genes were deleted and replaced by their human counterpart (similar by DNA sequence):

- Nearly half (47%) of the human genes tested, were able to rescue the loss of the yeast genes!
- This % was even higher when one matched human and yeast genes into gene modules (genes working together in the same biological process
- For e.g. in metabolic pathways like lipid, amino acid, carbohydrate and vitamin metabolism can be 80-90% replaceable)

- Kachroo et al. Science 2015. 348:921- 925

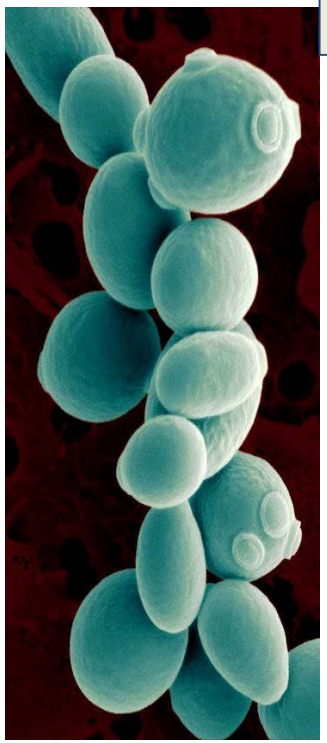
~ 1,000,000,000 Years of Evolution

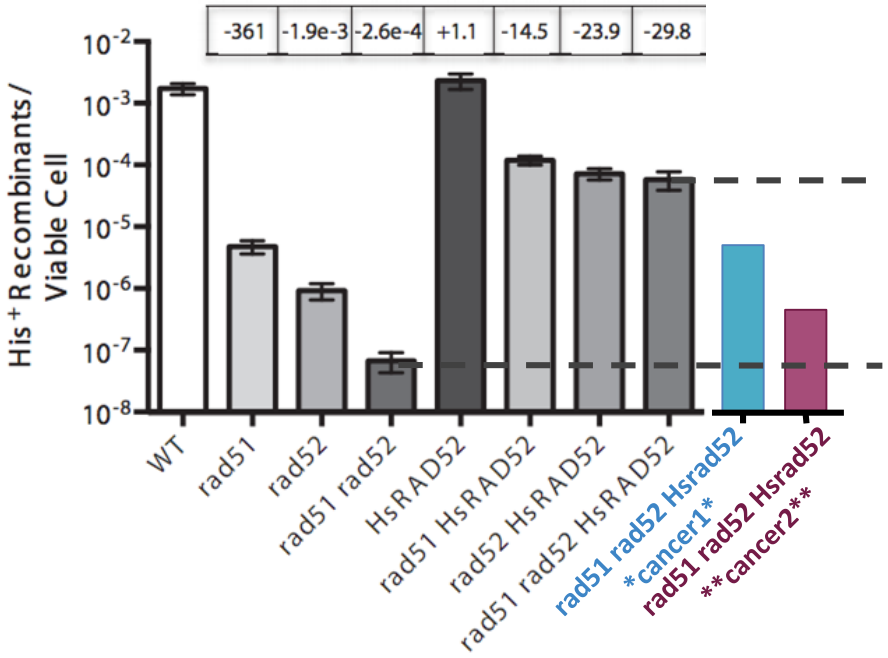
Modeling Cancer in Yeast and the Rad Genes



The breast cancer susceptibility genes BRCA1 & BRCA2 interact with Rad51.

All of these genes play a role in known human cancers





Manthey, Clear, Liddell, Negritto & Bailis. *Nucleic Acids Research,* 2016

What is BioSentinel?

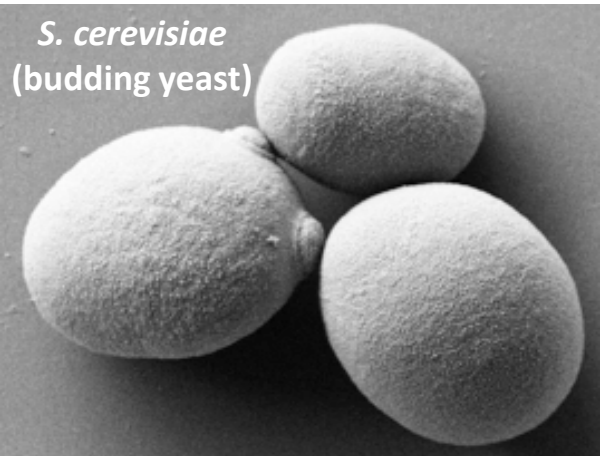
BioSentinel is a yeast radiation biosensor that will measure the DNA damage caused by space radiation, mainly DNA double strand breaks (DSBs).

Why?

Space radiation environment's unique spectrum cannot be duplicated on Earth. It includes high-energy particles, is omnidirectional, continuous, and of low flux. During solar particle events (SPEs), radiation flux can spike to a thousand times nominal levels.

How?

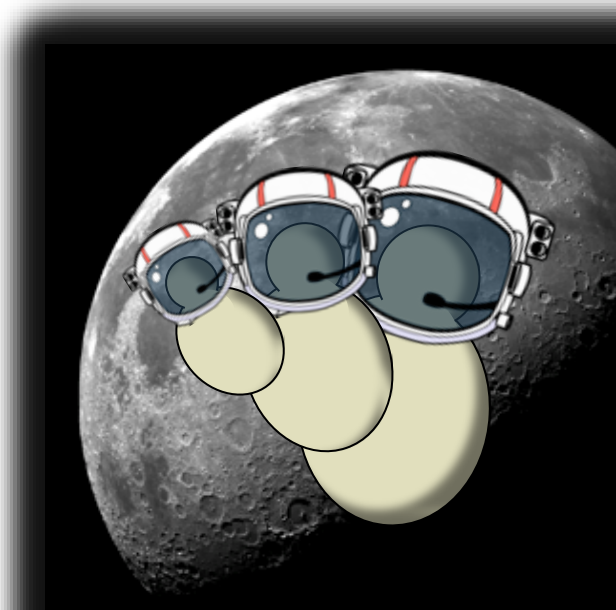
S. cerevisiae cells will sense & repair direct damage to their DNA. Yeast cells will remain dormant until nutrient media is added allowing cell growth in fluidic cards. Multiple cards will be in active mode during the mission & extra card(s) will be activated in the event of a Solar Particle Event (SPE).



Why budding yeast?

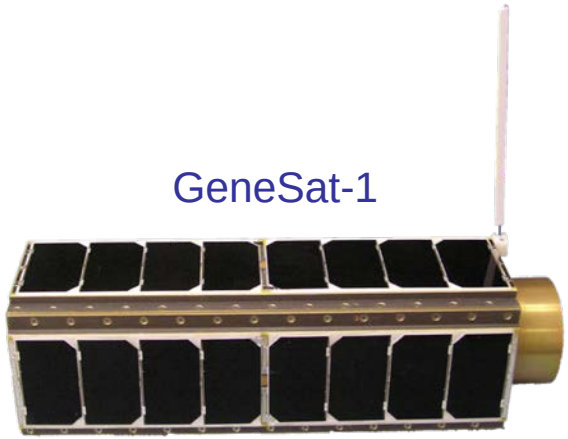
It is a eukaryotic cell (like human cells); easy genetic and physical manipulation; assay availability; flight heritage; ability to be stored in dormant state

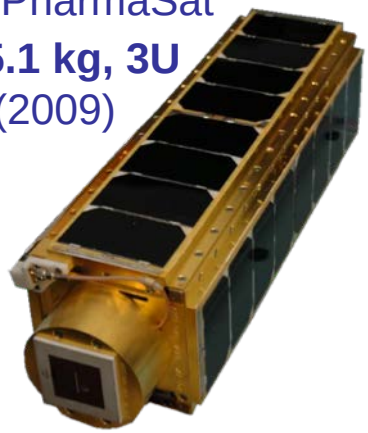
While it is a simple model organism, yeast cells are the best for the job given the limitations & constraints of spaceflight

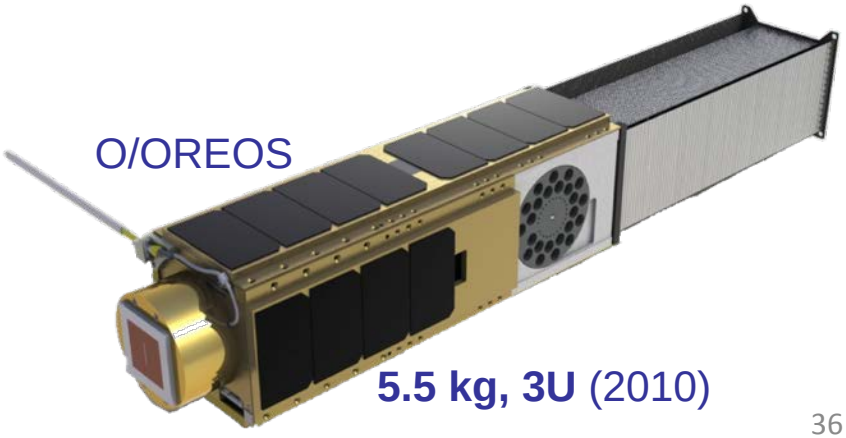


Rationale – Why Small Satellites?

- **Small Sats (< 50 kg) are ever more capable:** *Miniature/micro/nano technologies*
  - bioengineered organisms; (micro)fabrication; materials; optics; sensors; actuators; MEMS; fluidics; electronics; communications; instrumentation; data handling & storage
  - Power generation & storage density up; power consumption down
- **Access to space:** *Low-cost* launches as secondary payloads
  - *military, government, commercial*; US, Russia, Europe, Canada, India, …
  - ***Multiple flights possible*** - test, learn, iterate
- **Excellent education vehicle:** Significant academic participation worldwide
- **Autonomous operations:** Less reliance on human-tended experiments
- **Technology migration:** ISS; landers/orbiters for moon, Mars, other planets



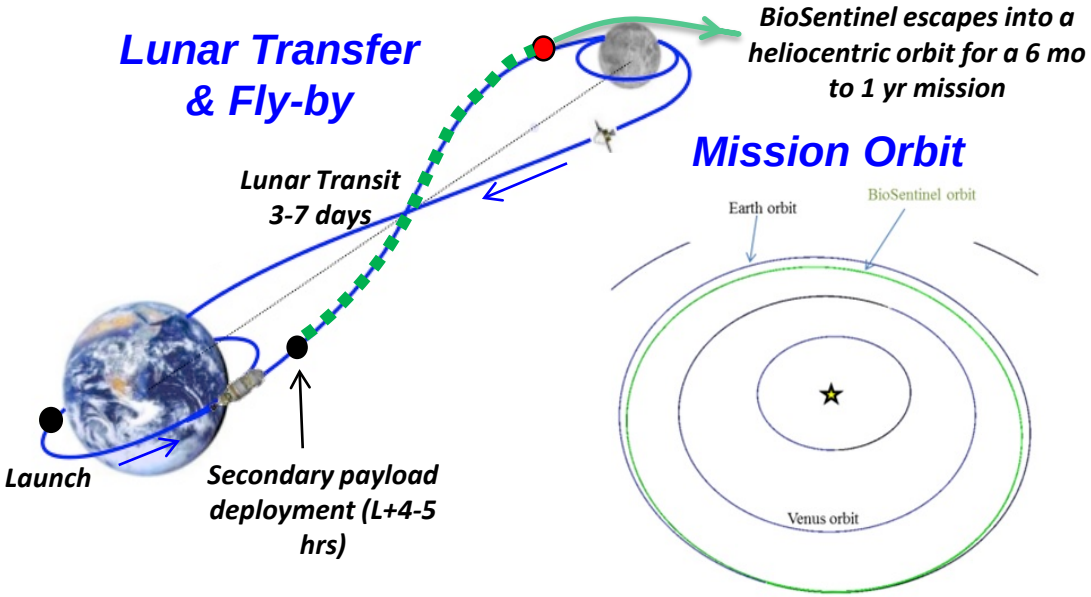




BioSentinel: Mission's firsts

- First biological study beyond low Earth orbit (LEO) in 40+ years
  - Secondary payload in SLS EM-1 (launch ~2018) - up to 13 CubeSats
  - Far beyond the protection of Earth's magnetosphere
  - BioSentinel will allow a comparison between different radiation & gravitational environments (interplanetary space, ISS, Moon, Mars, etc.)

- First biological 6U CubeSat to fly beyond LEO (~10 x 20 x 30 cm)
  - First CubeSat to combine biological studies with autonomous capability beyond LEO (autonomous response for both scientific measurements & response to SPE)
  - First mission to correlate simultaneous real-time biological and physical measurements of space radiation in an autonomous spacecraft



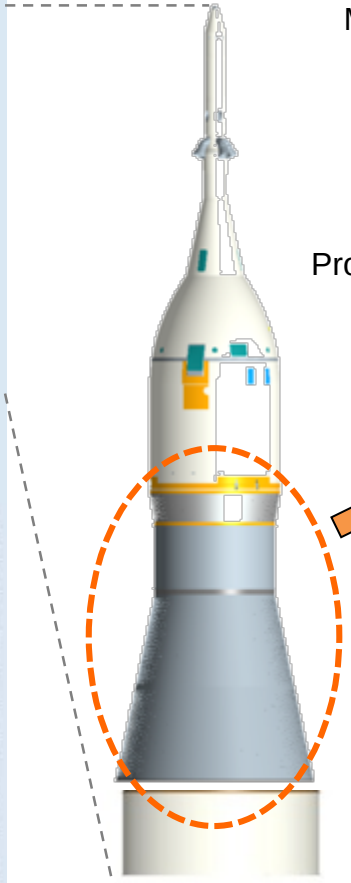
Distance to ISS: ~ 350 km

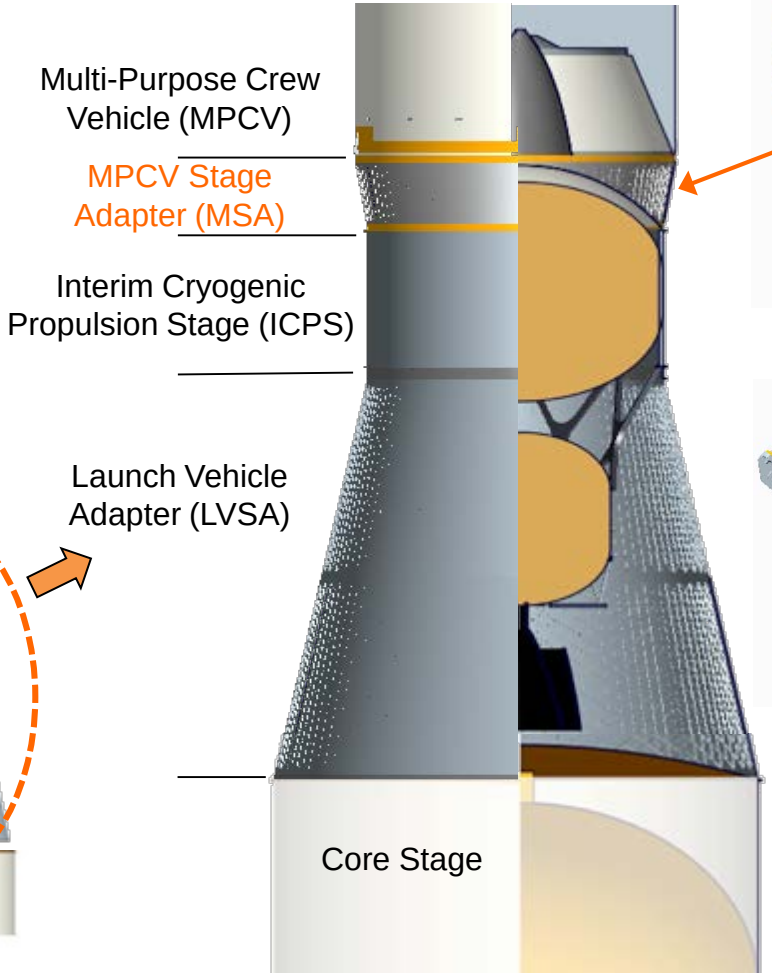
Distance to the Moon: ~385,000 km

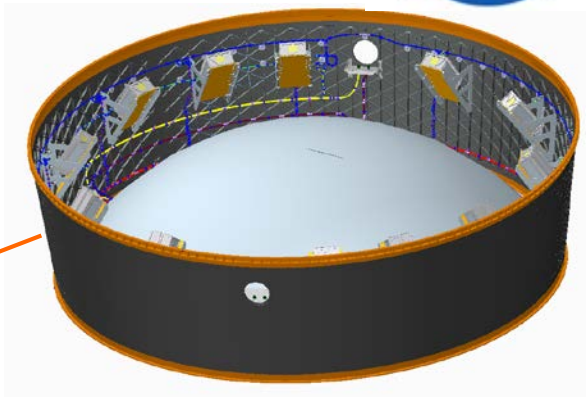
Distance to the Sun: ~150 million km

BioSentinel mission

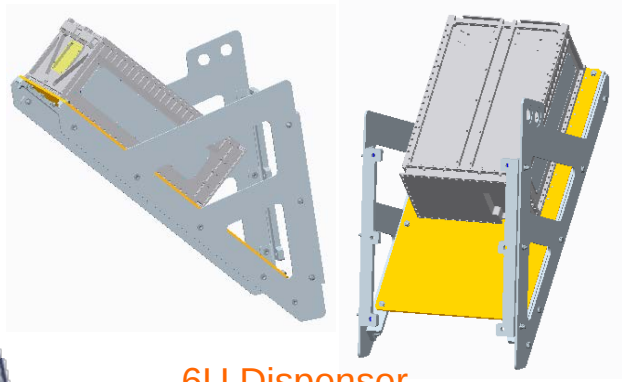
SLS EM-1



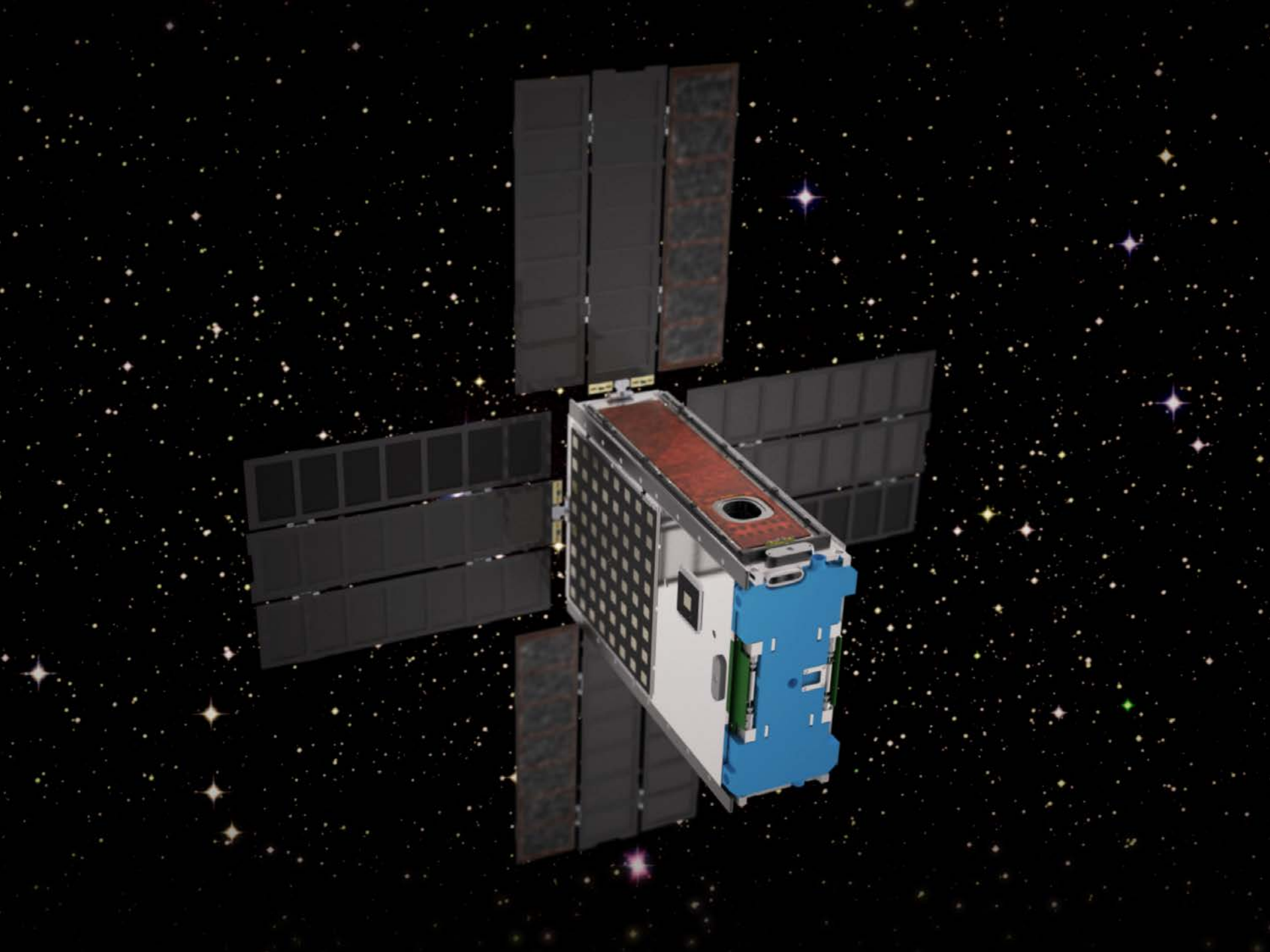




MSA Diaphragm

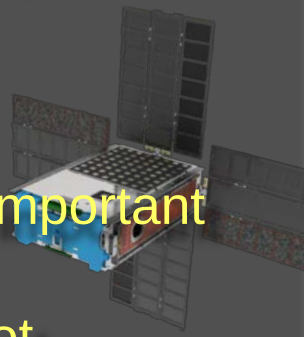


6U Dispenser



Conclusions

- **Biological studies in LEO show the importance of studying radiation effects in preparation for deep space human exploration**
- **Nanosats / cubesats can do real science in space!**
  - Tools/devices/methods of bio / nano / micro technologies are important enablers
  - Real-time, *in-situ* experiments provide insights on dynamics not available from expose-and-return strategies



- **Heritage of astro- & fundamental biology experiments in low Earth orbit is a major enabler for interplanetary biological missions**
  - Flying biology dry, filling fluidic µwells in µ-gravity
  - Long-term materials biocompatibility, stasis > 1 year, yeast in µgravity
  - Radiation-tolerant design: O/OREOS functional after ~ 5 years
  - Well-tested sensors, fluidic components & approach, optical measurement approach to growth & metabolism

BioSentinel (biosensor) team

Science - Sergio Santa Maria, Diana Marina, Macarena Parra, Tore Straume, Greg Nelson, Lauren Liddell, Sharmila Bhattacharya

Mission Management - Bob Hanel, Dawn McIntosh, James Chartres, Mario Perez, Elwood Agasid, Vas Manolescu, Matt D'Ortenzio

Payload - Charlie Friedericks, Rich Bielawski, Eric Tapio, Tony Ricco, Travis Boone, Ming Tan, Aaron Schooley, Mike Padgen, Lance Ellingson, Griffin McCutchenson, Diana Gentry, Dayne Kemp, Scott Wheeler, Susan Gavalas, Edward Semones

Spacecraft and Bus and Software - Hugo Sanchez, Matthew Sorgenfrei, Jesse Fusco, Vanessa Kuroda, Craig Pires, Shang Wu, Abe Rademacher, Josh Benton, Doug Forman, Ben Klamm

Collaborators: Loma Linda University, JSC Radworks, NASA Space Radiation Laboratory

Support: NASA Human Exploration and Operations Mission Directorate (HEOMD); Advanced Exploration Systems Division – Jitendra Joshi, Jason Crusan.