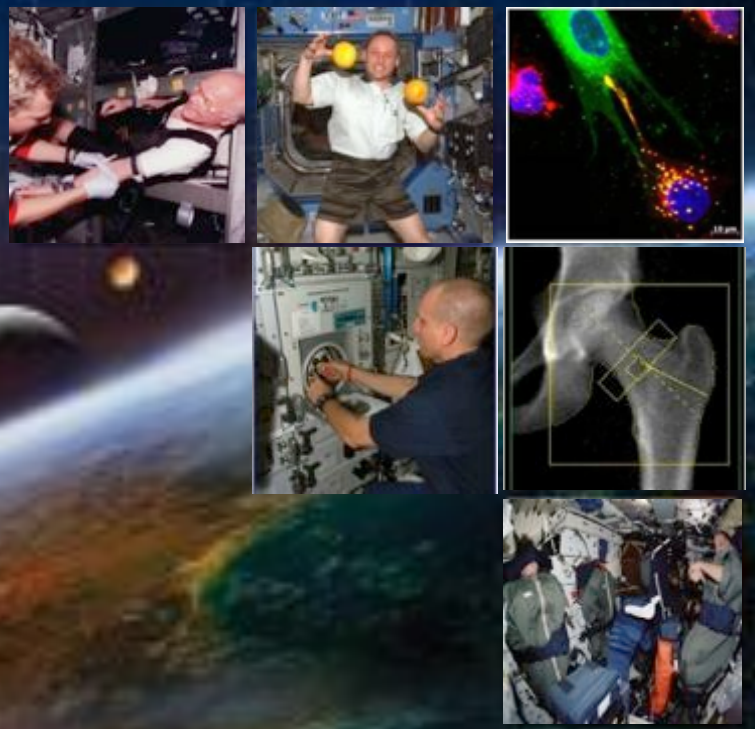
National Aeronautics and Space Administration

Human Research Program

The Twins Study: NASA's First Foray into 21st Century Omics Research

AsMA
14 May 2015



Craig E. Kundrot, Ph.D.
Deputy Chief Scientist, HRP
SA2/NASA JSC

The One-Year Mission

ISS Crew: Scott Kelly, Mikhail Kornienko Sign On For One-Year Mission

By: Tariq Malik
Published: 11/26/2012 08:12 AM EST on SPACE.com

A veteran NASA space commander and Russian cosmonaut have signed on for the ultimate space voyage: a yearlong trip on the International Space Station.

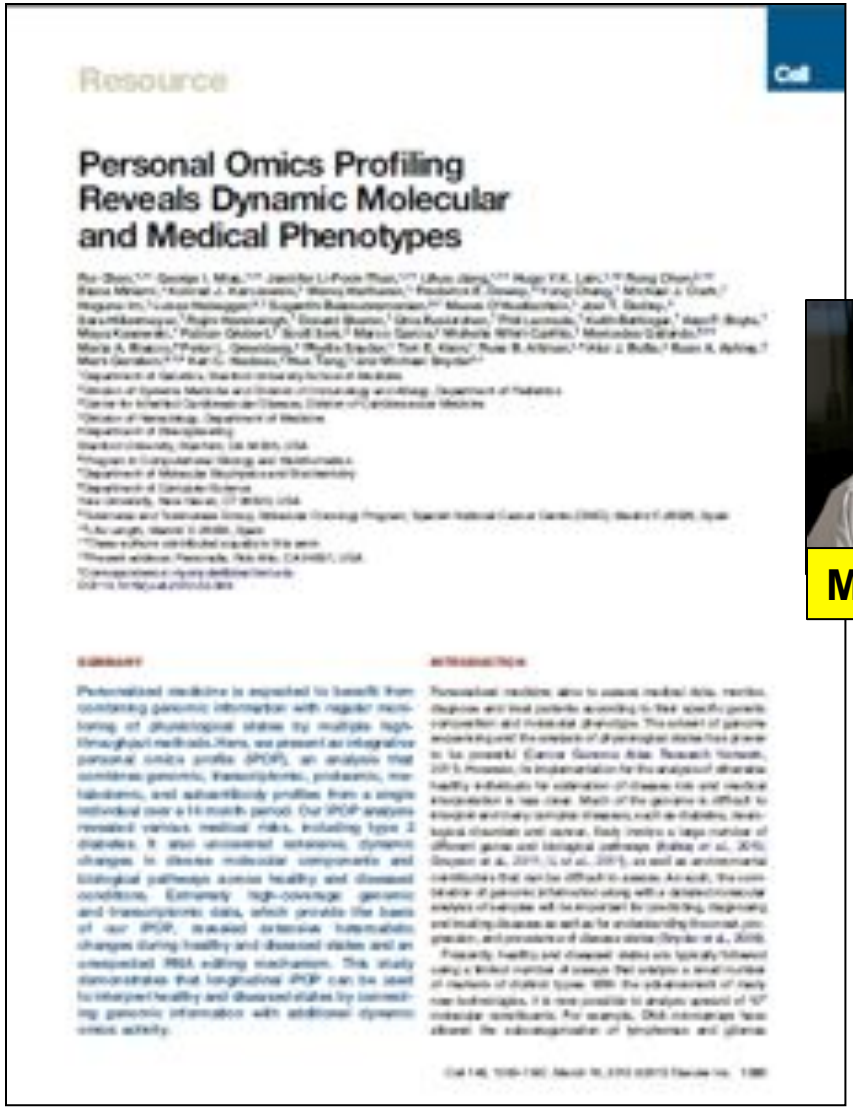
American astronaut Scott Kelly and Russian cosmonaut Mikhail Kornienko will launch on the one-year space station flight in spring 2015 and return to Earth in spring 2016, NASA officials announced today (Nov. 26). They will begin their mission training in early 2013.

The mission will help NASA understand how the human body adapts to extremely long space missions, such as voyages around the moon, to an asteroid and ultimately to Mars, NASA officials said.

Astronaut Scott Kelly Preparing for Unprecedented One Year in Space; Mission to Experiment on His Bone Mass, Vision, Immune System

A Chance in a Lifetime Opportunity for a Twins Study

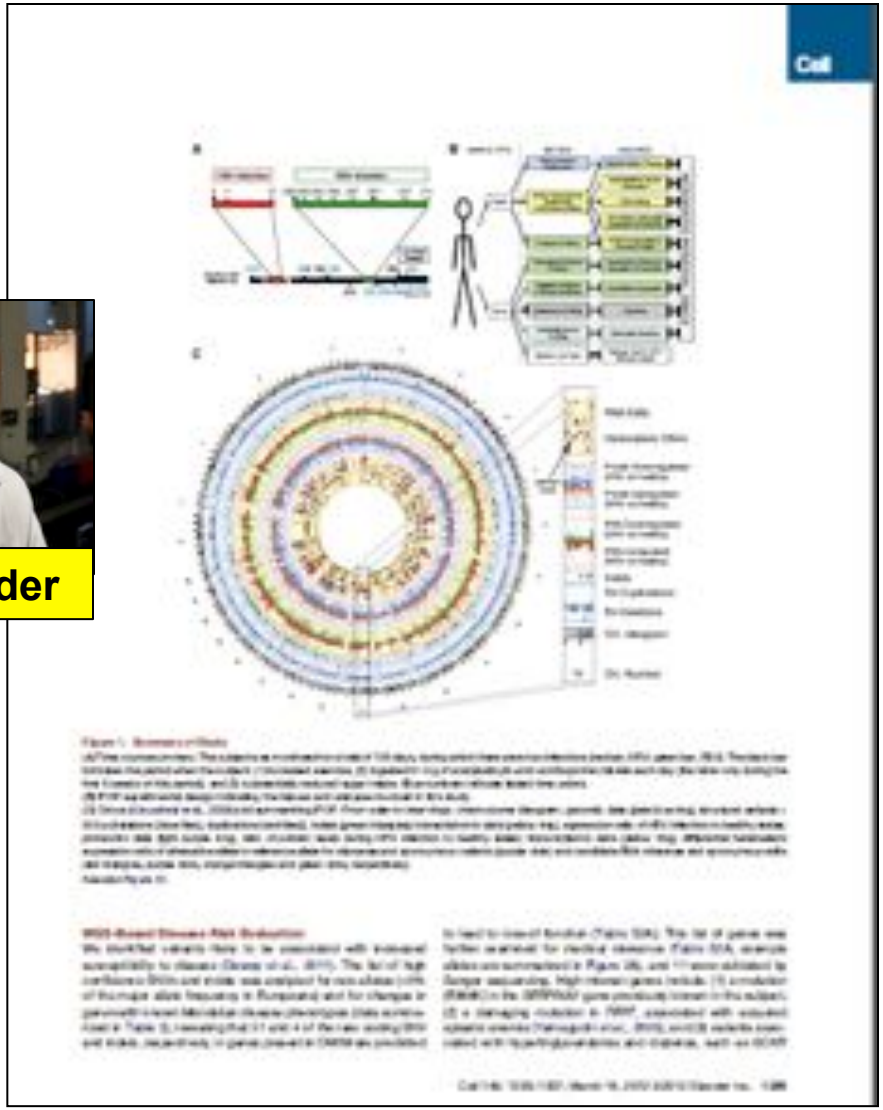
Great Value in n = 1 Omics Study Over Time


Resource

Cell

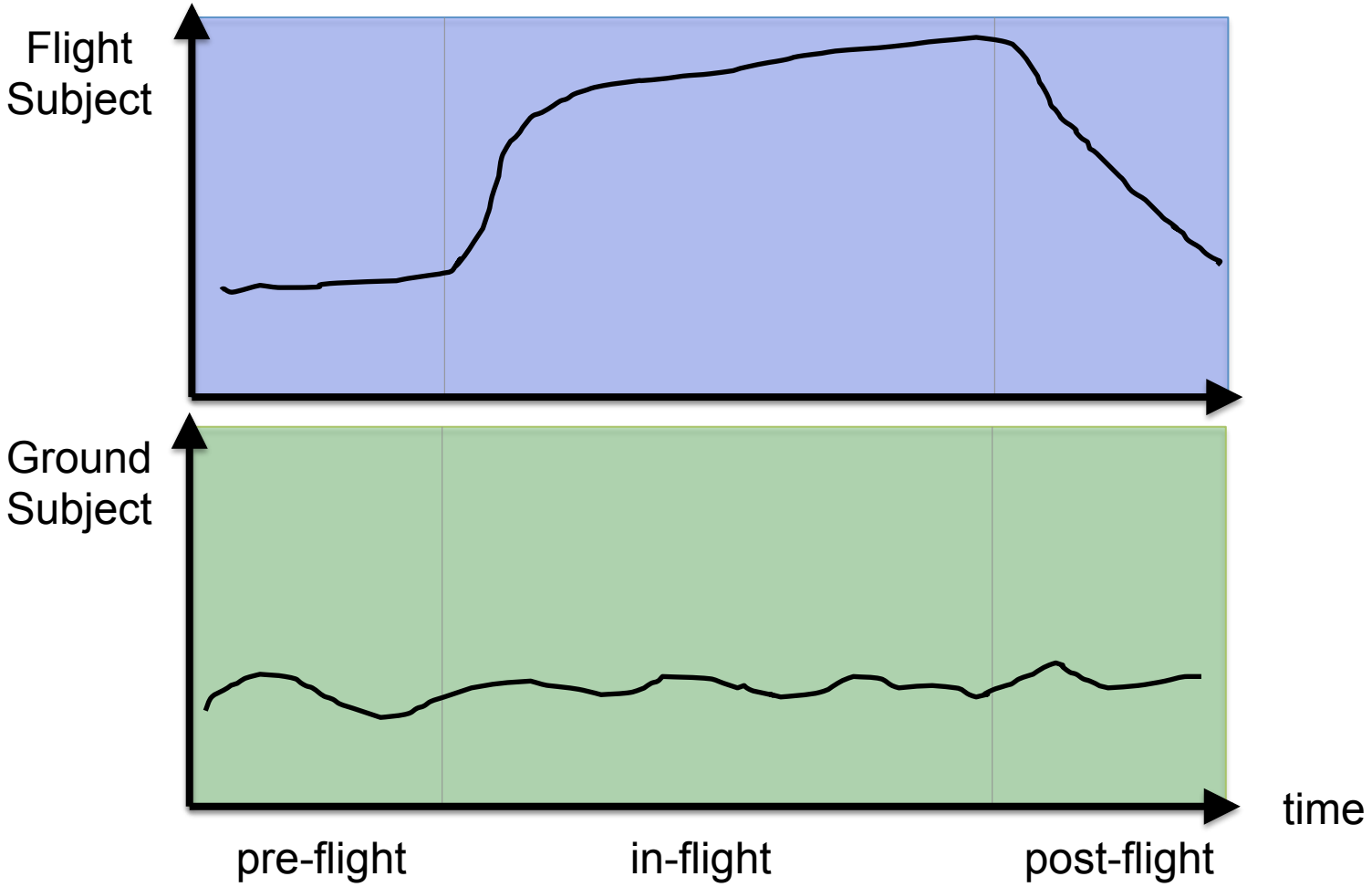
Personal Omics Profiling Reveals Dynamic Molecular and Medical Phenotypes


Mike Snyder

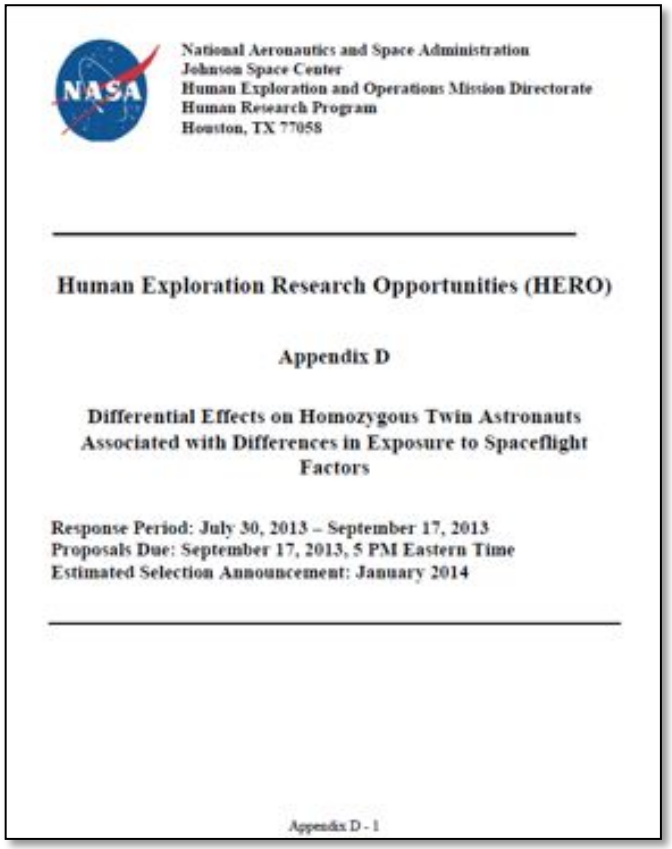

Cell

CellI (2012) 148:1293-1307.

One Notion


Flight Subject
Ground Subject
time
pre-flight
in-flight
post-flight

NRA Solicitation



National Aeronautics and Space Administration
Johnson Space Center
Human Exploration and Operations Mission Directorate
Human Research Program
Houston, TX 77058

Human Exploration Research Opportunities (HERO)

Appendix D

Differential Effects on Homozygous Twin Astronauts Associated with Differences in Exposure to Spaceflight Factors

Response Period: July 30, 2013 – September 17, 2013
Proposals Due: September 17, 2013, 5 PM Eastern Time
Estimated Selection Announcement: January 2014

Appendix D - 1

"To capitalize on this unique opportunity,

NASA's Human Research Program (HRP) and the National Space Biomedical Research Institute (NSBRI) are initiating

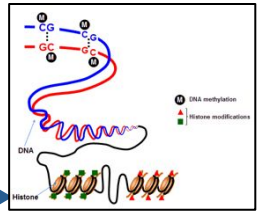
a *pilot demonstration project focused on the use of integrated human -omic analyses* to

better understand the biomolecular responses to

the physical,
physiological, and
environmental stressors

associated with spaceflight."

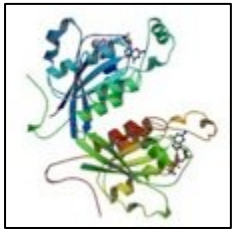
Buccal & Saliva



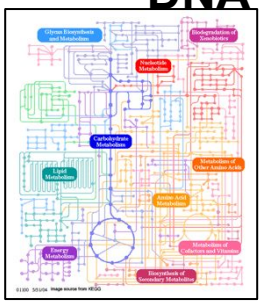
Epigenome

DNA

Urine

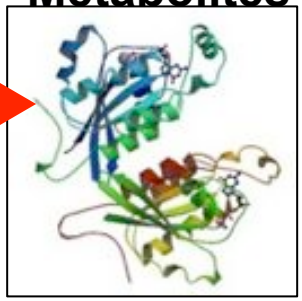


Proteins

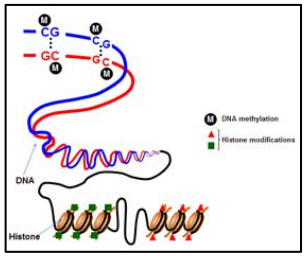


Metabolites

Blood

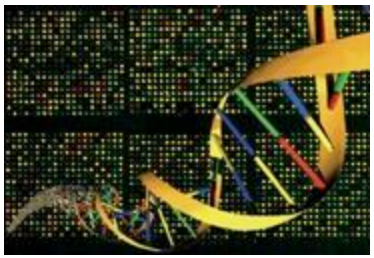


Proteins



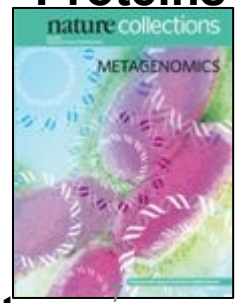
Epigenome

DNA



RNA

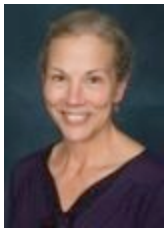
Stool



Metagenome

www.nasa.gov/content/twins-study

Differential effects on Telomeres and Telomerase in Twin astronauts associated with spaceflight



Susan Bailey, Ph.D.
Colorado State Univ

Kerry George
Wyle Labs/JSC

Specific Aims

The rate at which telomeres shorten provides an informative biomarker of aging and age-related pathologies (e.g., cardiovascular disease and cancer) that captures the interplay between genetics and lifestyle factors.

We propose that for the astronauts telomere maintenance is particularly relevant, as it reflects the combined exposures (e.g., radiation) and experiences (nutritional, psychological and physical stressors) encountered during space travel.

The Twins study provides the extraordinary opportunity to control variables of individual genetic differences, susceptibilities and lifestyle factors, making differential effects observed between the twins space-flight specific.

Comparisons with unrelated astronauts (separate study), will allow evaluating role of genetics/individual susceptibilities.

Implications of Research for Space & Earth

Space: This twins study will identify space-flight specific factors that influence telomere length and telomerase activity, informative biological indicators of aging and age-related degenerative diseases (e.g., cardiovascular disease and cancer). Our mechanistic investigations will begin to establish relevant relationships and suggest potential mitigation strategies for future study and to improve astronaut overall health.

Earth: Aging and age-related diseases like cardiovascular disease and cancer are an everyday concern on earth as well, therefore this study also seeks to make comparisons with unrelated astronauts (and controls) that will serve to identify individual susceptibility factors that influence telomere length and telomerase activity. Taken together with our mechanistic studies, mitigation strategies will be improved and applicable to all.

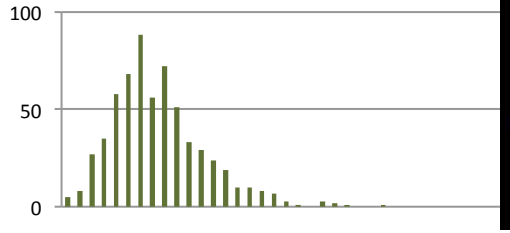
***Our goal* is to assess changes in telomere length and telomerase activity associated with the upcoming yearlong ISS mission in the space- and earth-bound twin astronauts.**

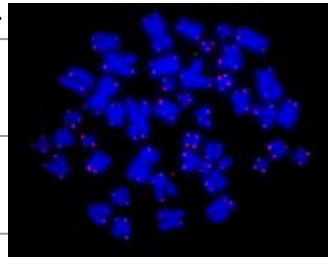
***We hypothesize* that accelerated telomere shortening and elevated telomerase activity will be associated with space flight** as compared to ground based control, in a duration and severity dependent manner.

- Blood samples will be taken ***pre-flight*** (to establish baseline), ***in-flight*** (to evaluate short-term/temporary changes) and ***post-flight*** (to evaluate long-term/permanent changes)
- Data sharing for other endpoints will also inform this effort
- *In vitro* studies will investigate potential mechanisms (e.g., oxidative stress) and mitigation strategies (e.g., antioxidants)

Telomere length will be assessed using TELO-FISH

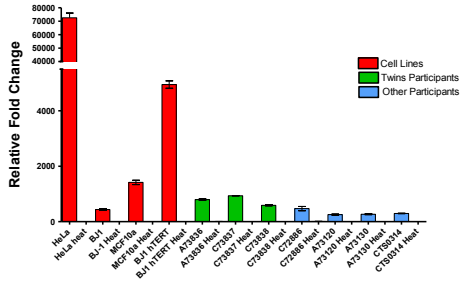
Florescence *in situ* Hybridization (FISH) with telomere probe on chromosomes (and interphase nuclei) is evaluated as Relative Fluorescence Intensity (RFI) distributions.





Telomerase activity will be assessed using qRT-PCR TRAP

quantitative Real Time-PCR Telomere Repeat Amplification Protocol



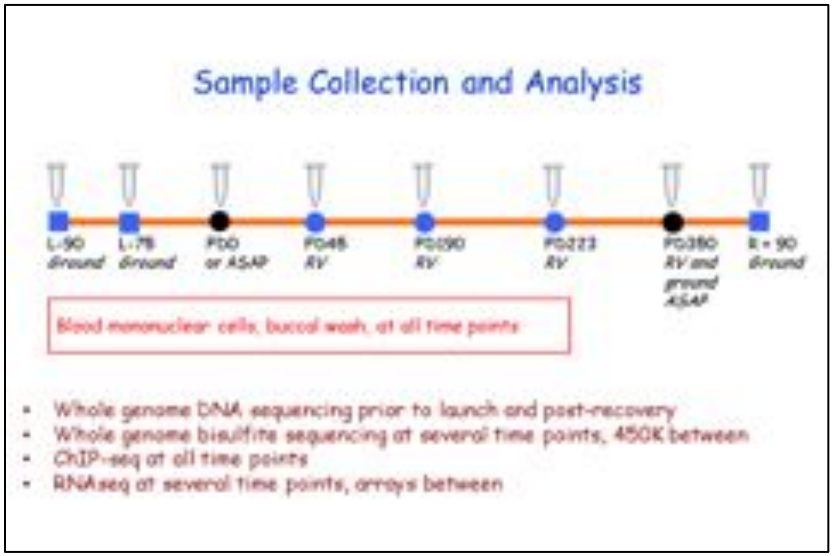
Epigenomics

Andrew Feinberg, M.D., M.P.H., and Jason Feinberg

Specific Aims

Aim 1. We will measure DNA methylation and chromatin at a genome-wide level in biological samples obtained from the space traveler at quarterly intervals, pre- and post-flight, and at times of unexpected exposures such as radiation events, or spacecraft environmental contamination. We also obtain measurements of the ground-based twin.

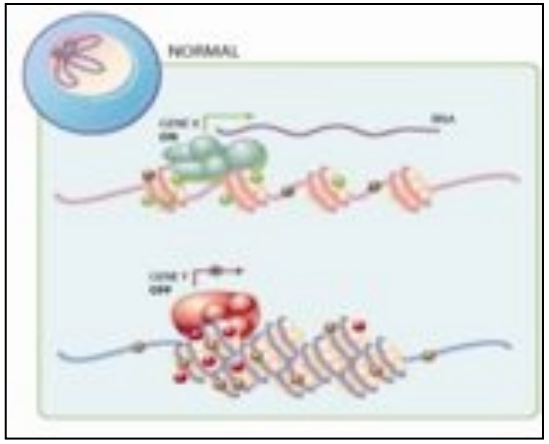
Aim 2. We will integrate epigenomic data with exposure to spaceflight conditions, looking for exposure-linked changes, and by comparison to the ground-based twin, determine whether these are transient or long-lived effects. We will also determine whether DNA mutations arise secondarily to these epigenetic changes.



Implications of the Research for Space & Earth

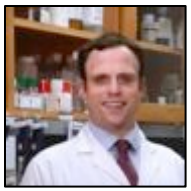
Space: Identify reversible causes of genomic damage in space, e.g. radiation or toxin induced epigenomics change; quantify aging and genomic exposure.

Earth: First human study of the epigenome over time in a defined/ controlled environment.



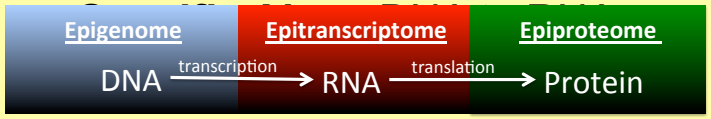
- DNA methylation
- Histone modifications (>200 known)
- Chromatin factor complexes
- Chromatin structure

Landscape of DNA and RNA Methylation



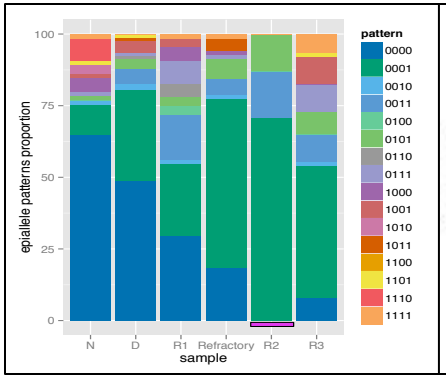
Christopher Mason, Ph.D.

Francine Garrett-Bakelman, M.D. Ph.D.



#1 – Genome-wide epigenetic profiles of DNA methylation changes
#2 – A comprehensive catalog of coding and noncoding, small and large RNA
#3 – Transcriptome-wide maps of RNA methylation sites

Δ in Epigenetics : Loci, regions, and clones

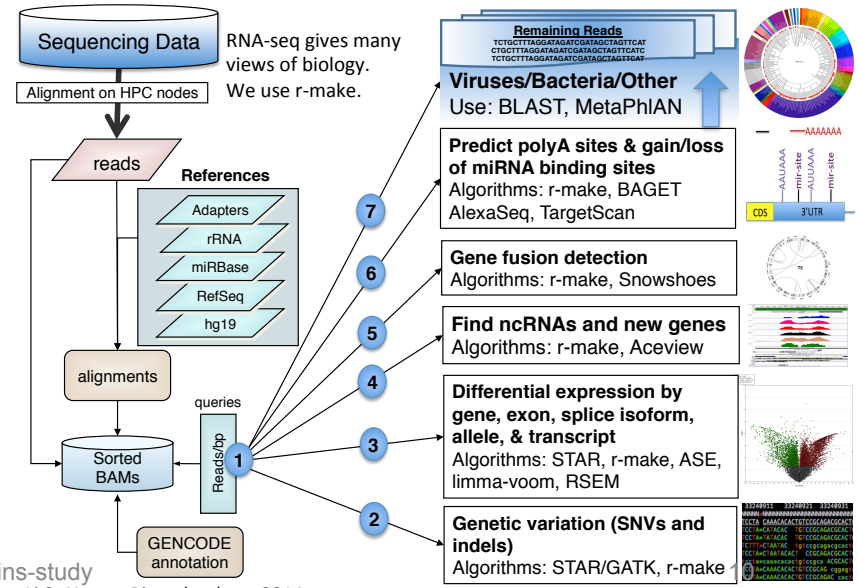


Δ in Transcriptome : Genes, Isoform, Edits, Allele, SNVs, ncRNAs, Fusions, & Methylation

Implications of the Research for Space & Earth

Space: (1) Establish the genetic networks and expression patterns activated by space travel, (2) trace clonality of epigenetic changes, (3) examine the methylation of RNA

Earth: Aid research on aging, cancer, RNA biology, and circadian rhythm, all of which show differences at the (epi)genome & (epi)transcriptome



Li S, *Nature Biotechnology*, 2014.

Biochemical Profile:
Homozygous Twin control for a 12 month Space Flight Exposure

Scott M. Smith, Ph.D.

Specific Aims
To provide a database of biochemical analyses from blood and urine samples. The analyses reflect a broad set of nutritional and physiological variables that may be altered as a result of the space flight environment (including diet, stress, weightlessness). Collecting data on the Ground twin will allow for a more direct comparison of the effects of space flight on human biochemistry and physiology.

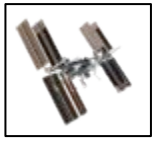
Blood and urine collections

Preflight:
L-180, L-45, L-10

In-flight:
FD15, 30, 60, 120, 180, 240, 300, 360

Post flight:
R+0, R+30

Implications of the Research for Space & Earth



Space:
Improve understanding and time course of biochemical changes during flight and how the changes relate to diet during flight.

Earth:
Improve understanding of how diet can impact different biological systems.

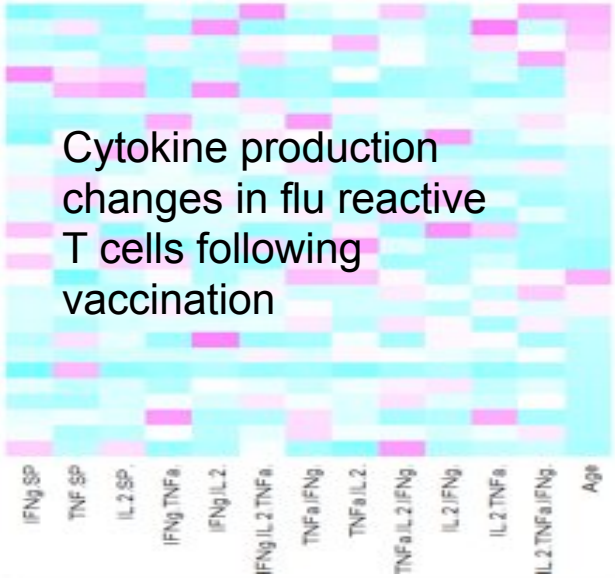
Immunome studies in space

Emmanuel Mignot, MD, PhD

Stanford University

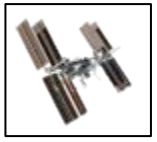
Specific Aims

- Study how long term space travel affects the immune system
- We will study how parameters of the immune system change at baseline and after a seasonal flu vaccination
- To do so, we study baseline and post flu parameters before, during and after a one year space flight



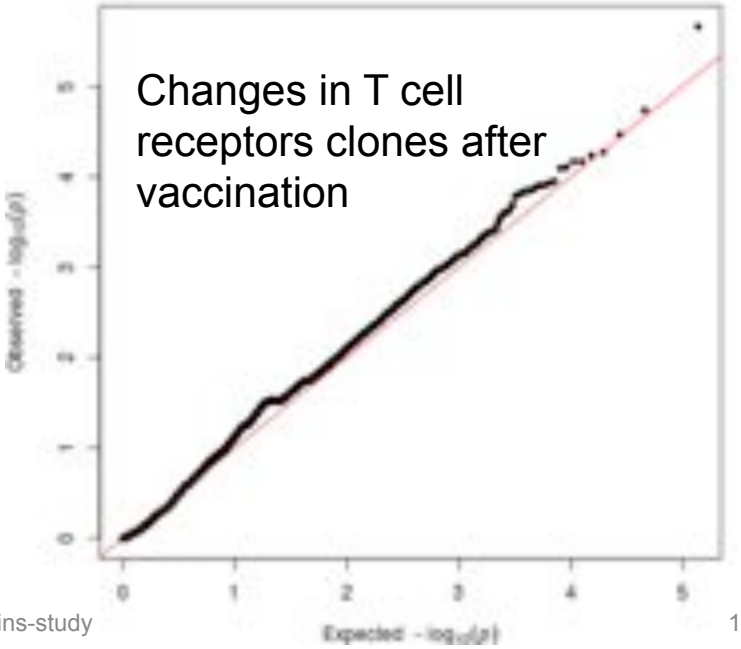
Cytokine production changes in flu reactive T cells following vaccination

Implications of the Research for Space & Earth



Space:
Will ensure that astronauts keep a healthy immune system during long flight, and stay protected against infections from earth when visitors are coming.

Earth:
Understand how immune response to vaccination differ in twins



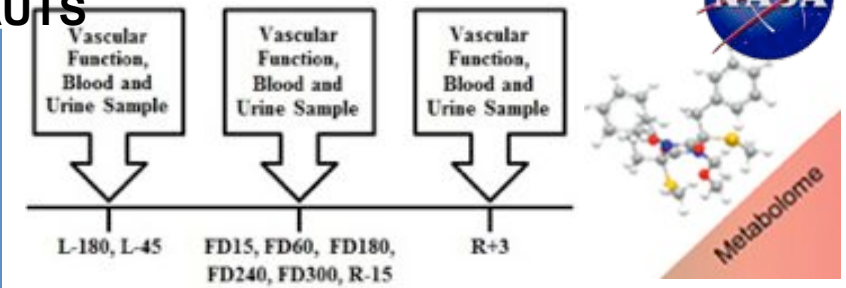
Changes in T cell receptors clones after vaccination

METABOLOMIC AND GENOMIC MARKERS OF ATHEROSCLEROSIS IN TWIN ASTRONAUTS

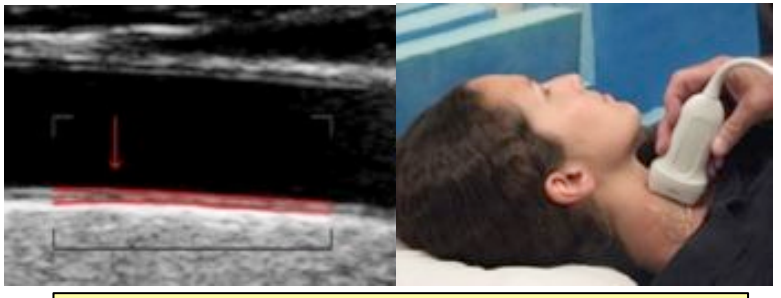
Stuart Lee

Specific Aims

- To study the effects of long-duration spaceflight on the cardiovascular system independent of genotype
- To investigate relationships between gene expression, metabolomic profiles, biomarkers in blood and urine, and arterial structure and function using the space-flown and the ground-based identical twin



Pre- and Post-flight Testing



Implications of the Research for Space & Earth

Space: Determine if the spaceflight environment perturbs genomic and metabolomic profiles and accelerates development of atherosclerosis (occupational health)

Earth: Develop novel insights of how longitudinal changes in genomic and metabolomic profiles are related to risk factors for atherosclerosis

Inflight Operations

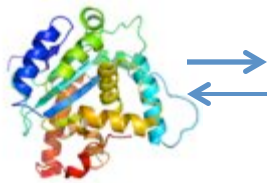
PROTEOMIC ASSESSMENT OF FLUID SHIFTS AND ASSOCIATION WITH VISUAL IMPAIRMENTS AND INTRACRANIAL PRESSURE IN TWIN ASTRONAUTS

Brinda Rana, PhD
Mike Stenger, PhD
Vivian Hook, PhD

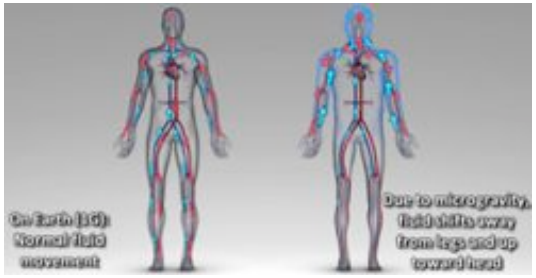
Specific Aims

To explore proteomic and genomic biomarkers underlying space flight-induced fluid shifts and visual impairment & intracranial pressure (VIIP) symptoms.

The proteome is the set of proteins produced by the genome at a given time. Proteomics captures the state of molecular and cellular processes at a specific time point.



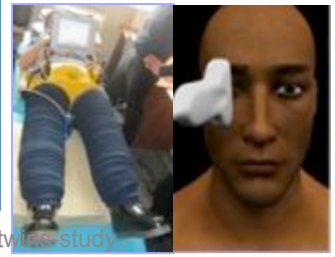
Blood Plasma proteins



In-flight Operations

Blood Plasma collection
Ultrasound measures of fluid shifts
Intracranial Pressure
Intraocular Pressure
Ocular Structure
Blood Pressure
Heart Rate
Vascular Resistance

Pre- and Post-flight Testing



All in-flight operations and:
Tissue hydration
MRI

Implications of the Research for Space & Earth

Space: Discovery of molecular pathways involved in the evolution of spaceflight adaptations related to fluid redistribution in-flight and the etiology of visual acuity and ocular changes in-fight and post-flight.

Earth: This project has broader impact on Earth-based clinical areas such as traumatic brain injury-induced elevations of intracranial pressure, hydrocephalus, and glaucoma

Cognitive Performance in Spaceflight

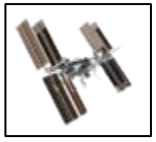
Mathias Basner, M.D., Ph.D.

Ruben C. Gur, Ph.D.

Specific Aims

There are a number of environmental stressors unique to the spaceflight environment that may affect cognitive performance, which is crucial for mission success. Our main objective in the TWINS study is to investigate whether cognitive performance is affected by initial and prolonged exposure to the spaceflight environemnt and after return to Earth. We will use the *Cognition* test battery, which consists of 10 brief neuropsychological tests that were specifically designed for high performing astronauts. We will compare data within subjects, between twins, relative to astronauts flying 6-month missions, and relative to normative data gathered in astronauts on the ground. The cognitive data will be correlated with markers derived from biological samples taken before, during, and after the 12-month mission.

Implications of the Research for Space & Earth



Space: Exploration-type missions will require humans to spend unprecedented durations in space, yet our knowledge on the effects of prolonged exposure to the spaceflight environment is very limited. After the study, we will have an initial understanding of whether and to what extend prolonged ISS missions are associated with changes in cognitive performance, and how these relate to biologic markers.

Earth: The results have direct implication for other high performing populations exposed to stressful environments for prolonged periods of time on Earth.

	Test	Cognitive Domain	Brain Regions (from fMRI studies)	Avg. Time (Min)
	Motor Praxis (MPT)	Sensory-motor ability	Sensorimotor Cortex	0.51
	Visual Object Learning (VOLT)	Visual object learning and memory	Medial Temporal Cortex - Hippocampus	1.69
	Fractal 2-Back (F2B)	Attention and working memory	Dorsolateral prefrontal Cortex, Cingulate, Hippocampus	1.93
	Abstract Matching Task (AMT)	Abstraction and mental flexibility	Prefrontal Cortex	2.33
	Line Orientation (LOT)	Spatial orientation	Right Temporo-Parietal Cortex, Visual Cortex	2.07
	Emotion Recognition (ERT)	Emotion recognition	Cingulate Cortex, Amygdala, Hippocampus, Fusiform Face Area	2.03
	Matrix Reasoning (MRT)	Abstract reasoning	Prefrontal Cortex, Parietal Cortex, Temporal Cortex	2.09
	Digit Symbol Substitution (DSST)	Complex scanning, visual tracking, attention	Temporal Cortex, Prefrontal Cortex, Motor Cortex	1.60
	Balloon Analog Risk (BART)	Risk decision making	Orbital frontal Cortex, Amygdala, Hippocampus, Anterior Cingulate Cortex	2.39
	Psychomotor Vigilance (PVT)	Vigilant attention and psychomotor speed	Prefrontal Cortex, Motor Cortex, Visual Cortex	3.17

The Cognition Test Battery

Cognition was specifically designed for astronauts and is currently used during 6-month ISS missions and in multiple space analog environments (including Antarctica, HI-SEAS, and HERA).

The Bacteriome in the Gastrointestinal Tract

Fred Turek, Ph.D.

Specific Aims

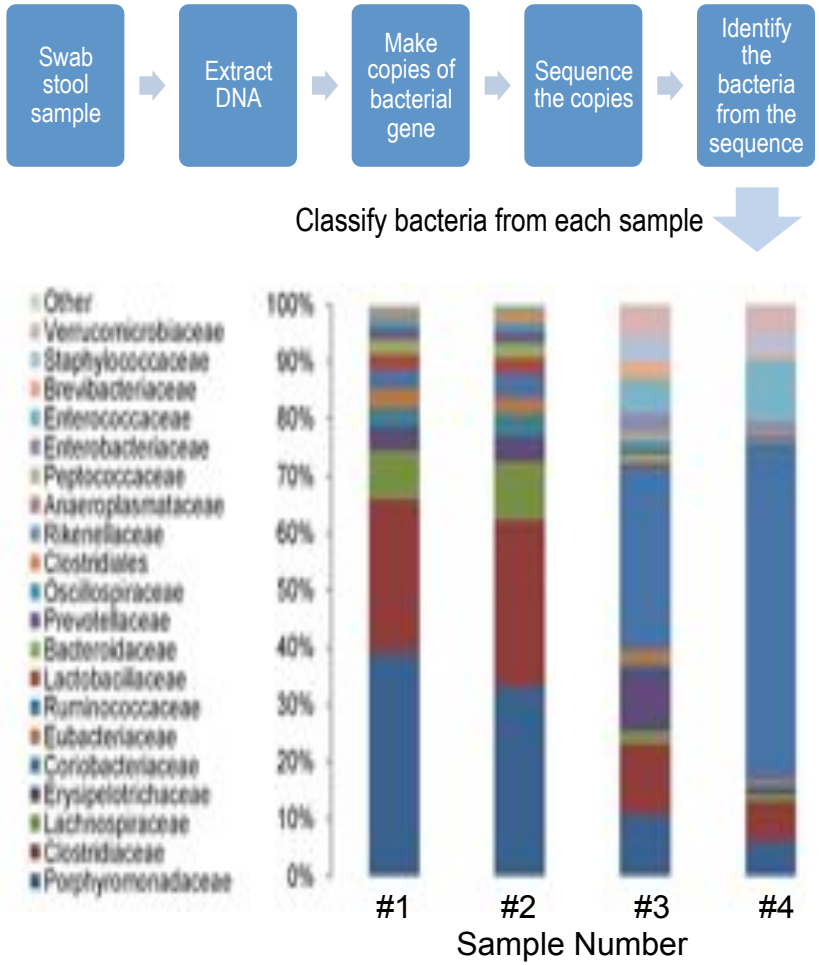
The GI tract of humans is populated by a diverse "ecosystem" of micro organisms, mostly bacteria: the bacteriome. The bacteriome can help-- contributing to digestion and immune system function-- or harm-- overgrowth of some types accompanies illness or stress.

This project will examine what changes occur to the bacterial populations over a year in space, that are different from the changes over time on Earth. Are particular types of bacteria susceptible to the space environment, and if so, which types?

Implications of the Research for Space & Earth

Space: Knowing how the bacteriome changes over time in space can help us make plans to protect astronauts' health for longer-term space flights. For example, adjustments to diet could help maintain beneficial bacterial types.

Earth: Observing how the bacteriome changes in relation to health and environmental changes, (such as those studied in other Twin Projects) can provide insights into how the bacteriome may respond to challenges and contribute to the human host's health.



Relative abundance of different families of bacteria. Will there be systematic changes in the twin in space not seen in the twin on Earth?

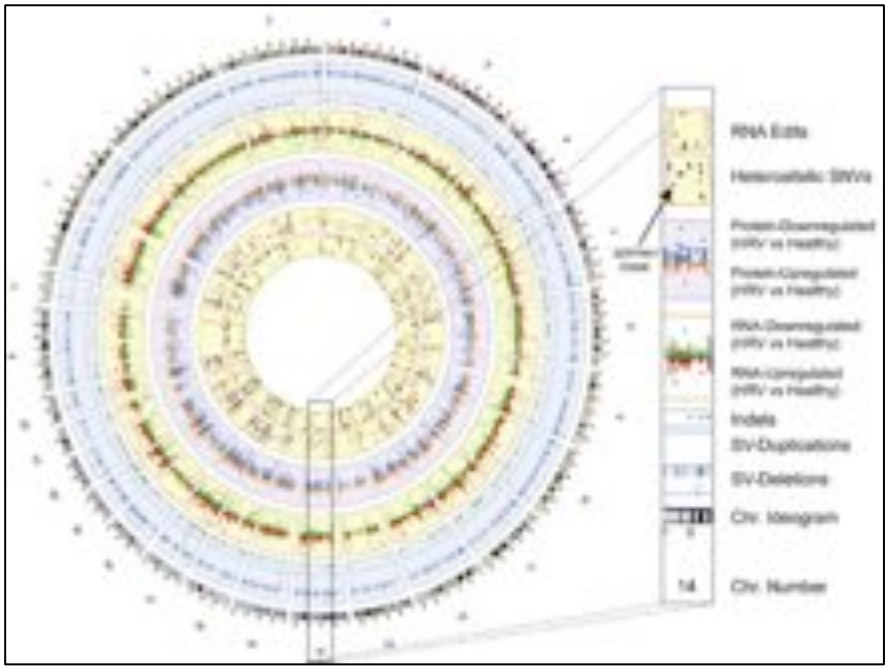
Integrated Omics: Mike Snyder, Ph.D.

Michael Snyder, Ph.D.

Juliane Winkelmann, M.D.

Specific Aims

Our main objective in the twin study is to perform a complete analysis of all biomedical and molecular data collected during the mission to produce the singular most comprehensive portrait of the human biophysical response to the rigors of spaceflight. We are at an unprecedented era in genomic medicine, allowing for the sensitive and precise measurement of billions of biochemical molecules, which will allow us to detect the subtlest of changes in Scott and Mark's physiology over time. By integrating these data, we can follow alterations in their cellular systems to both better understand the effects of space travel on human health, and how an astronaut's genome may contribute to his/her own unique physiologic response to microgravity.

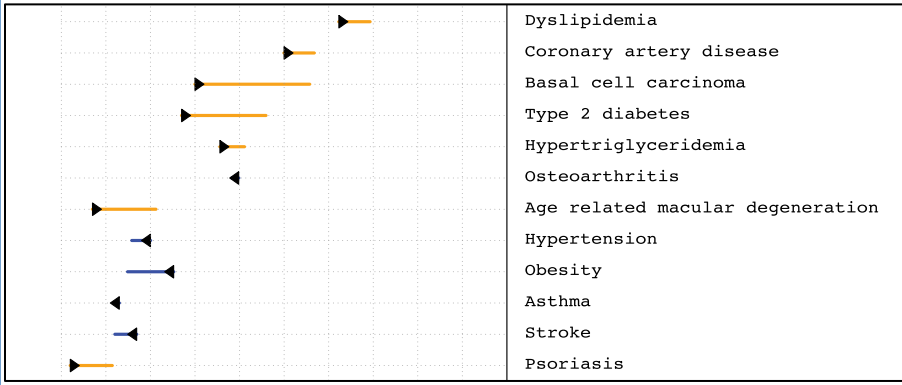


Integrative multi-omic model

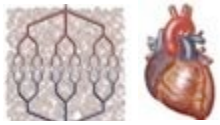
Implications of the Research for Space & Earth

Space: We will generate a detailed benchmark for how human physiology changes in space in great molecular detail. This wealth of data will be essential for any future planning of long duration space exploration missions, and provide a proof-of-principle for better monitoring and managing astronaut health.

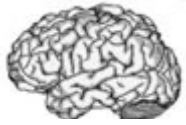
Earth: With this study, Scott and Mark Kelly will be the most thoroughly profiled twins in history, and the resultant data will offer new insights into how two siblings with nearly-identical genomes respond to different conditions.



Risk-o-gram

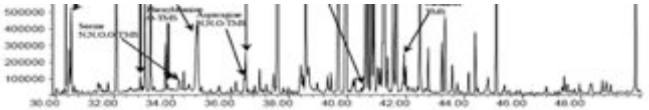


Vasculature
Lee

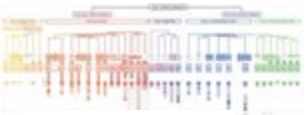


Cognition
Basner

Microbiome
Turek



Targeted and Global Metabolomics
Lee/Rana, Mignot/Snyder & Smith

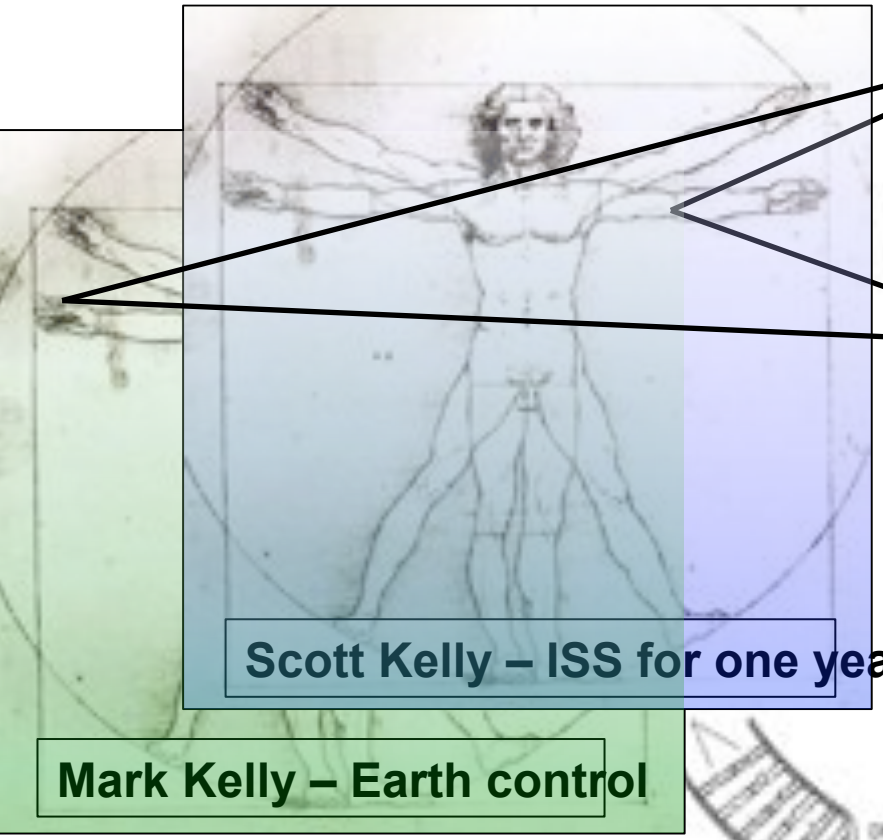


Cytokines
Mignot

Telomere Length
Bailey

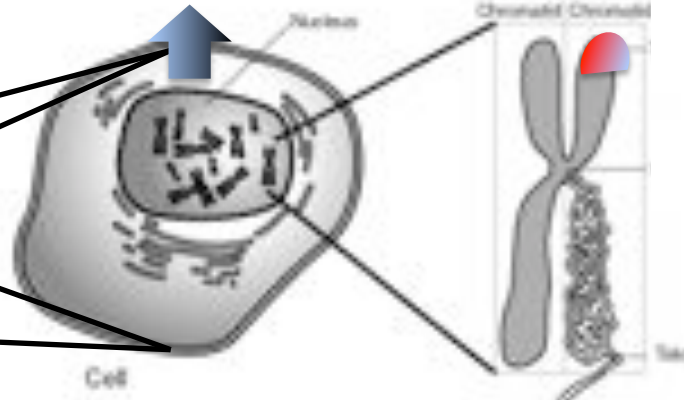
B-cells / T-cells
Mignot

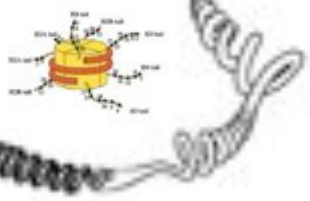
Antibodies
Mignot/Snyder



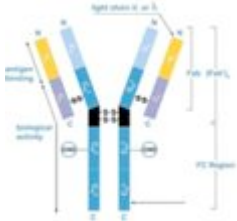
Scott Kelly – ISS for one year

Mark Kelly – Earth control



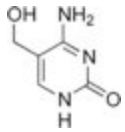


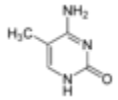
Chromatin
Feinberg



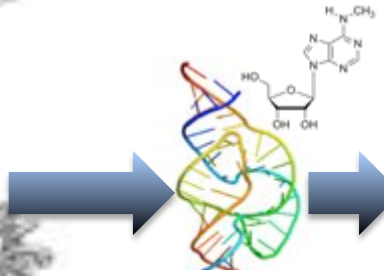
DNA Mutations
Feinberg

DNA Hydroxy-methylation
Mason

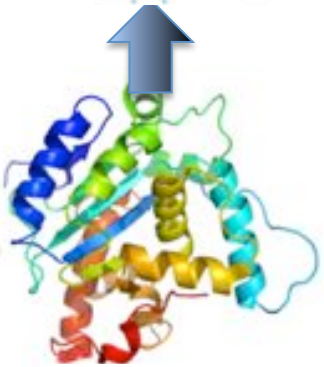




DNA Methylation
Feinberg & Mason

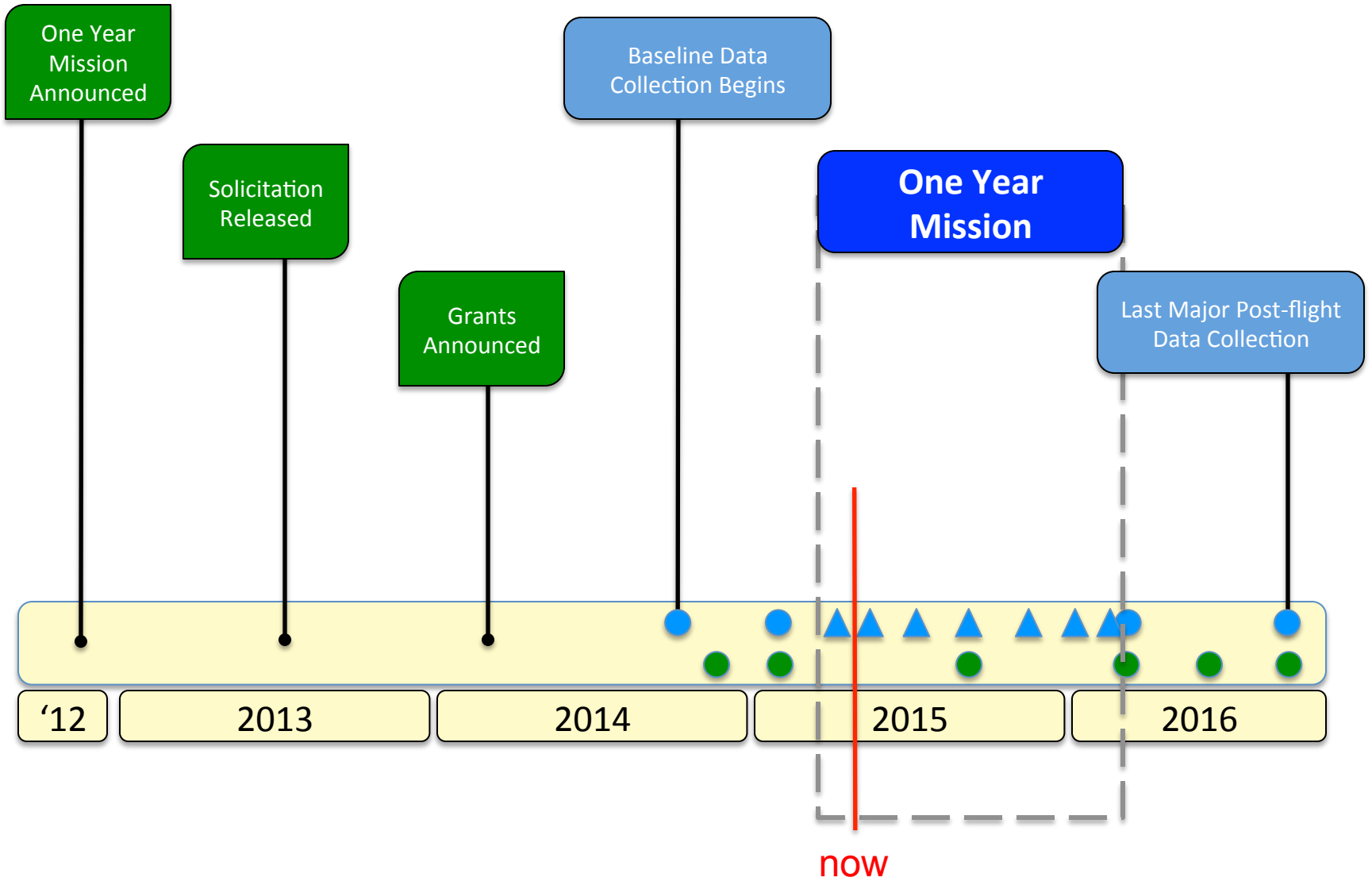


large/small RNA
& RNA Methylation
Mason



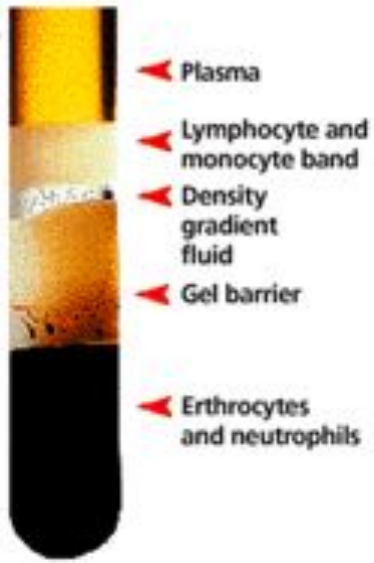
Proteomics
Lee/Rana

Timeline


One Year Mission Announced
Solicitation Released
Grants Announced
Baseline Data Collection Begins
One Year Mission
Last Major Post-flight Data Collection
'12
2013
2014
2015
2016
now

Progress to Date

- Pre-flight, baseline data collections complete
  - Most protocols already established
  - New protocols developed for Cell Preparation Tubes (CPTs)
    - To sort or not to sort?



- In-flight collections have begun
  - CPTs processed and frozen
  - CPTs processed and en route to Houston right now!

Issues Associated with Omic Research

- Research ethics
  - The primary risks involved in genetic research are risks of social and psychological harm, rather than risks of physical injury
    - Could provoke anxiety and confusion about disease risk
    - Uncover unwanted information about heritage, ancestry, and family relationships
  - De-identification of genomic information is difficult
    - Astronauts are public figures
  - Information given to subjects
    - Individual genome sequence data?
    - Interpretation of the genome sequence and/or genetic counseling?
    - Option to decline to receive all or part of the results (Right Not to Know)?
  - Research community's access to genomic information
  - Interim policy on genetic research JID 1800.4
  - NASA-wide policy under development

- Medical care
- Occupational health
- Insurance (health, disability, life)
- Employment activity

Conclusion

- The Twins Study (Scott and Mark Kelly) is NASA's first foray into 21st-century omics research
  - Built around Scott Kelly's one year mission
- The Twins Study will examine
  - Genome, telomeres, epigenome
  - Transcriptome and epitranscriptome
  - Proteome
  - Metabolome
  - Physiology
  - Cognition
  - Microbiome
- NASA is addressing
  - Protections for research participants
  - Use of data in medical care, occupational medicine, mission planning

Acknowledgements

The Twins Study Investigator Team

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Mike Barratt

Bill Paloski

Graham Scott

Jeff Sutton

Mark Shelhamer

Acknowledgements

time.com/meet-the-twins-unlocking-the-secrets-of-space/

National Aeronautics and Space Administration