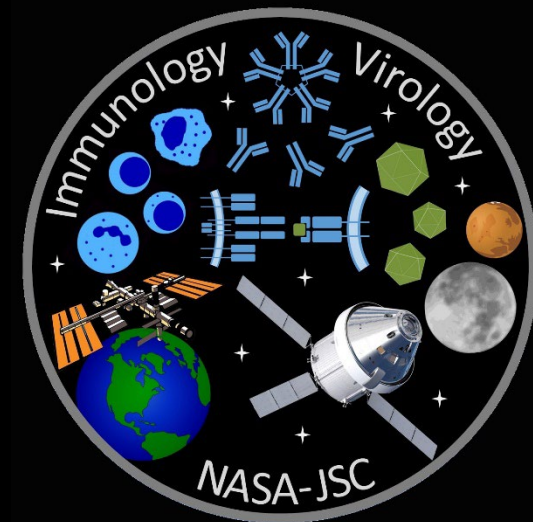


Monitoring of Deep Space Physiology via Dry Saliva Sampling during Artemis Missions (*Artemis Biomarkers*) – Project Update



Cody Gutierrez¹, Douglass Diak², Sara Bustos-Lopez¹, Mayra Nelman-Gonzales³, Satish Mehta³, Scott M. Smith⁴, Sara R. Zawart⁵, Richard Simpson⁶, Grace M. McKenzie⁶, Alexander Chouker⁷, Brian Ctucian⁴

¹JES Tech, ²Aegis Aerospace, ³KBR, ⁴NASA Johnson Space Center, ⁵The University of Texas Medical Branch at Galveston,

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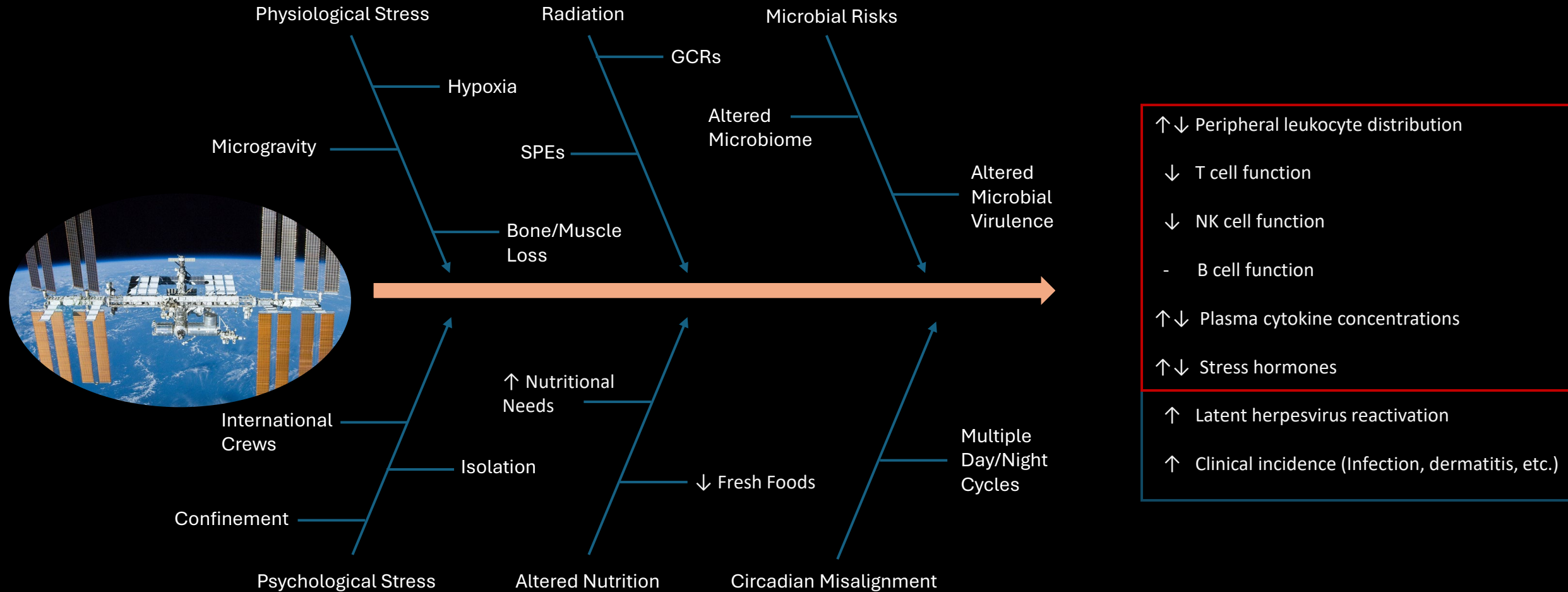
Spaceflight and Health



- Fluid Redistribution to upper body
- Otoliths in inner ear respond differently, eyes become main way to sense motion
- Plasma volume decreases, anemia
- Elevated radiation may increase cancer risk
- Dysregulation of the immune system
- Muscle and bone weakening; bone demineralization
- Elevated kidney stone risk



Spaceflight Immune System Dysregulation



Clinical Relevance

International Journal of General Medicine

Dovepress

open access to scientific and medical research

ORIGINAL RESEARCH

Incidence of clinical symptoms during long-duration orbital spaceflight

Crucian et al.; International Journal of General Medicine 2016;9: 383–391.

viruses

MDPI

Article

Dermatitis during Spaceflight Associated with HSV-1 Reactivation

Satish K. Mehta ^{1,*}, Moriah L. Szpara ², Bridgette V. Rooney ³, Douglass M. Diak ⁴, Mackenzie M. Shipley ², Daniel W. Renner ², Stephanie S. Krieger ⁵, Mayra A. Nelman-Gonzalez ⁵, Sara R. Zwart ⁶, Scott M. Smith ⁷ and Brian E. Crucian ⁷

Mehta et al.; Viruses 2022, 14, 789. <https://doi.org/10.3390/v14040789>

Case report

Immune system dysregulation preceding a case of laboratory-confirmed zoster/dermatitis on board the International Space Station

Satish K. Mehta, PhD,^a Rahul Suresh, MD,^b Keith Brandt, MD,^c Douglass M. Diak, PhD,^d Scott M. Smith, PhD,^b Sara R. Zwart, PhD,^e Grace Douglas, PhD,^b Mayra Nelman-Gonzalez, BA,^b Simon Clemett, PhD,^f Tyson Brunstetter, OD, PhD,^b and Brian E. Crucian, PhD^b

Mehta et al.; J. Allergy Clin. Immunol. Global – 2024; 3:100244.

Clinical Communications

A case of persistent skin rash and rhinitis with immune system dysregulation onboard the International Space Station

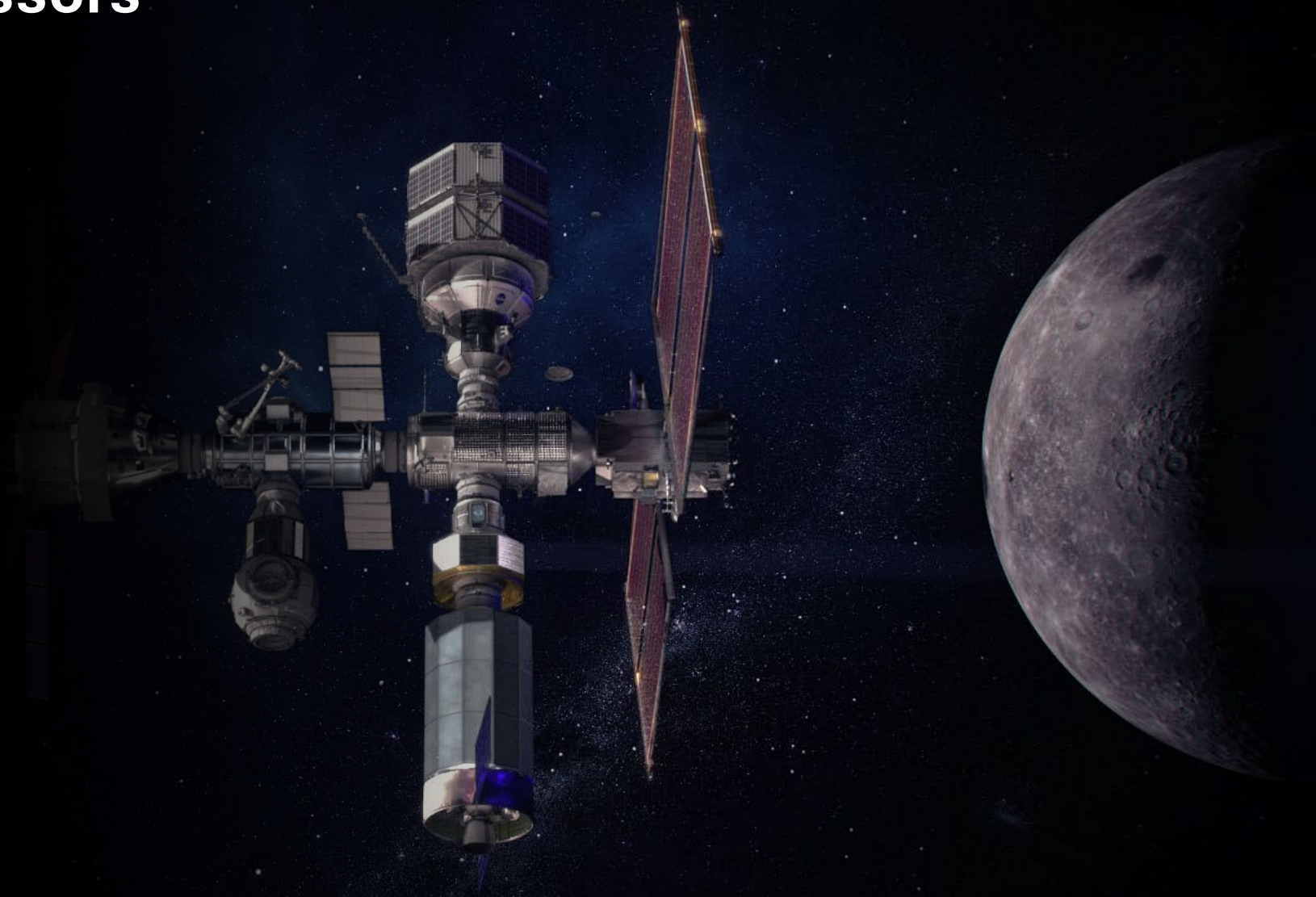
Brian Crucian, PhD^a, Smith Johnston, MD^b, Satish Mehta, PhD^c, Raymond Stowe, PhD^d, Peter Uchakin, PhD^e, Heather Quiriarte, BA^c, Duane Pierson, PhD^g, Mark L. Laudenslager, PhD^f, and Clarence Sams, PhD^b

Crucian et al.; J Allergy Clin Immunol Pract. 2016 Jul-Aug;4(4):759-762.e8.

Medical Conditions	Total events	Events/ person year
Allergic Reaction	1	0.06
Anaphylaxis	0	---
Upper Respiratory Infection (combination of rhinitis, nasal stuffiness and sneezing)	5	0.301
Eye Infection	0	---
Herpes Zoster	5	0.301
Otitis Media/Externa (ear pain, or ear stuffiness+congestion)	17	1.022
Pharyngitis (sore throat)	1	0.06
Sepsis	0	---
Sinus Infection	0	---
Skin Infection (including scalp pruritis, pus forming wounds on wrist, finger)	5	0.301
Skin Rash/Hypersensitivity (including skin conditions such as tinea versicolor, dermatitis, rosacea)	23	1.383
Urinary Tract Infection	1	0.06
Malignancies*	0	---
Autoimmunity*	0	---
Infections, Other* [#]	11	0.666
Total:	69	4.18

Deep Space Stressors

- ↑ Radiation
- ↑ Stress
- ↓ Exercise
- Hypoxia/Hyperoxia
- ↓ Frequent resupply
- Diet
- BHP support
- Return option
- Clinical management



Study Aims

- Monitoring astronaut physiology via analysis of bio samples is well established. The *Immunology Biomarkers* study will monitor physiological adaptation to deep space in Artemis crewmembers.
- Samples will be collected Pre/In/Post-Mission. The returned data will span immunology, inflammation, stress, nutritional status and radiation damage.
- Small pocket-sized booklet with several specialized pages will be utilized to collect samples in space, which will allow for long term non-conditioned storage.
- Each crewmember deposits saliva on their own booklet at predetermined timepoints. The book is then stowed until return.

Why Artemis/Lunar Mission?

- On Lunar missions, all mission stressors will be increased (radiation, isolation, habitable volume, etc.), and our ability to care for the crew will be decreased (limited power/volume, no rapid return option, etc.).
- Prior to prolonged deep space missions of exploration, initial Artemis/Lunar missions can be used to determine the effect of deep space travel on physiology and therefore influence medical/countermeasure decisions to protect future astronauts.
- Dry saliva and fingerstick blood will enable monitoring of physiology to potentially occur on Lunar habitats.

Dry Technology: Biomarkers and Virus Books

5x Saliva pages total separated by wax sheets.
Collected 1x time per collection TP.

Artemis
Dry Saliva Book

**Biomarkers
Book**

(5 Pages)

Collect all pages on
the same day at the
same time

Name: _____

Date/Time: _____

Wax Paper

Saliva Page

Wax Paper

ARTEMIS
DRY SALIVA BOOK
'BIO-MARKERS'
(4 pages)

Collect all pages on
the same day at the
same time

NAME: _____

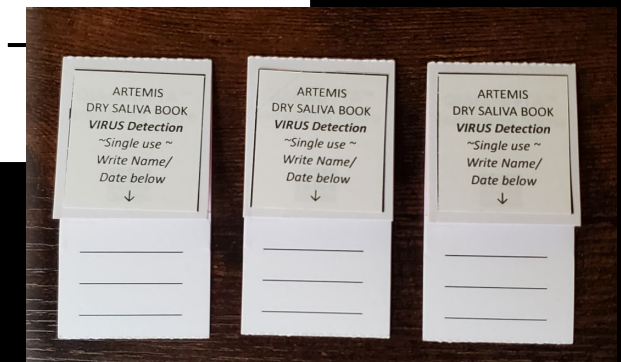
DATE/TIME of
COLLECTION: _____

1x Pre-treated saliva page collected.
Collected 3x times over 3 days per collection TP.

Artemis
Dry Saliva Book
**Virus
Detection**
~Single Use~

Write Name/
Date Below

Saliva



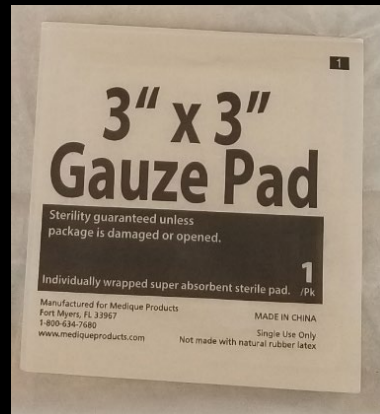
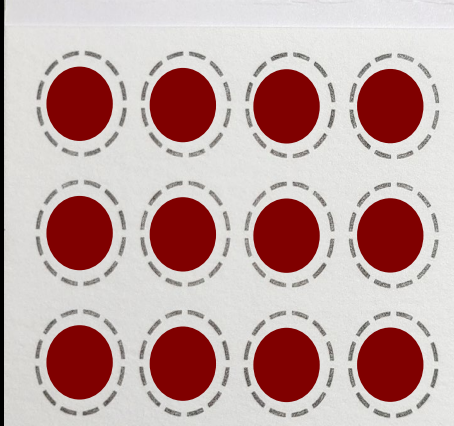
Dry Saliva Book Analytes

Virus Book	Biomarkers Book						
Virus DNA	Stress Hormones	Immuno-Regulation		General Immune	Anti-Microbial	Nutrition	Radiation
EBV	Cortisol	IL-1a	IL-15	CRP	Defensins	Ferritin/Heme	BDNF
VZV	DHEA	IL-1b	IP-10	IgA	HNP 1-3	Angiotensin II	
HSV-1		TNFa	IL-13				
		IL-6	IL-9				
		IL-1ra	FGF-2				
		IL-8	Flt-3L				
		IL-2	Fractalkine				
		INFy	GRO				
		IL-4	IFN-a2	IgG	Lysozyme	OPG	
		IL-5	MCP-3				
		IL-10	MDC				
		IL-13	PDGF-AA				
TTV		IL-17	PDGF-AB/BB	IgM	LL37	RANKL	
		MCP-1	TGFa				
		MIP-1a	TNFB				
		MIP-1b	sCD40L				
		RANTES	MIG				
		G-CSF	IL-17E/IL25				
		GM-CSF	LI-17F				
		VEGF	IL-18				
		IL-7	IL-22				
		EGF	IL-27				
		EOTAXIN	M-CSF				
		IL-12(p40)					
		IL-12(p70)					

*Assay additions may occur, in the areas of clinical chemistry, nutritional status, bone, radiation, or immunobiology, pending further stability studies

Dry Technology: Dry Blood Card and Analytes

12x Blood spots are collected via a fingerstick.
Collected 1x time per collection TP.



Dry Blood Card			
Clinical Chemistry		Immuno-Regulatory	
hsCRP	TPOab	IL-1a	IL-15
Vitamin D2	Thyroglobulin	IL-1b	IP-10
Vitamin D3	Cortisol	TNFa	IL-13
Estradiol	SHBG	IL-6	IL-9
Testosterone	PSA	IL-1ra	FGF-2
Progesterone	LH	IL-8	Flt-3L
Cortisol	FSH	IL-2	Fractalkine
DHEAs	Copper	INFy	GRO
Cholesterol	Zinc	IL-4	IFN-a2
Triglycerides	Selenium	IL-5	MCP-3
HDL	Magnesium	IL-10	MDC
HbA1C	Cadmium	IL-13	PDGF-AA
Free T3	Lead	IL-17	PDGF-AB/BB
Free T4	Mercury	MCP-1	TGFa
Total T4	AMH	MIP-1a	TNFb
TSH	Insulin	MIP-1b	sCD40L
		RANTES	MIG
		G-CSF	IL-17E/IL25
		GM-CSF	LI-17F
		VEGF	IL-18
		IL-7	IL-22
		EGF	IL-27
		EOTAXIN	M-CSF
		IL-12(p40)	
		IL-12(p70)	

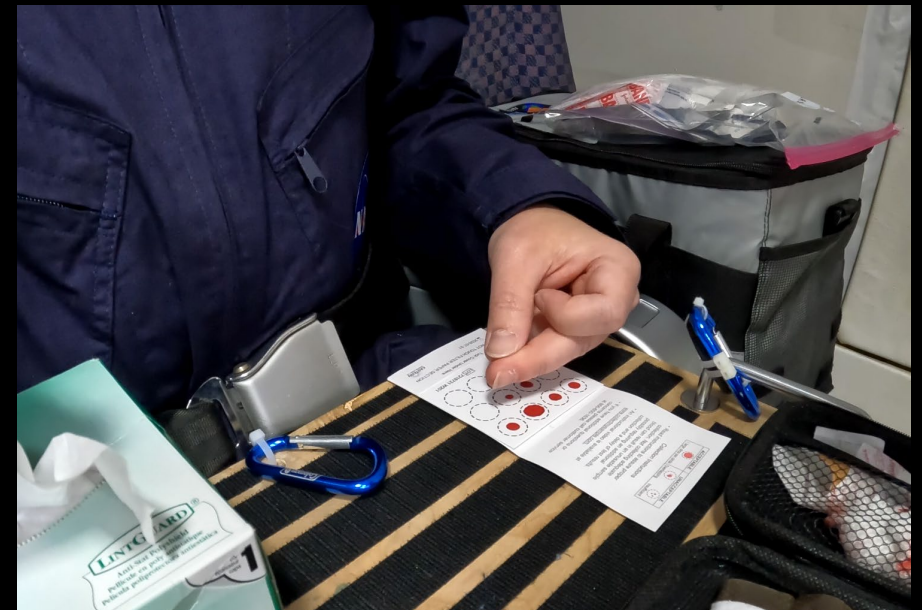
Dry Technology: Validation

Microgravity/Parabolic Flight Validation

Verified:

1. Hardware compatibility w/ microgravity
2. Feasibility of collection
3. Collection method/protocol for crewmembers

A prototype of the biomarkers book was Flown on the ISS, which allowed for the Dry collection of cortisol



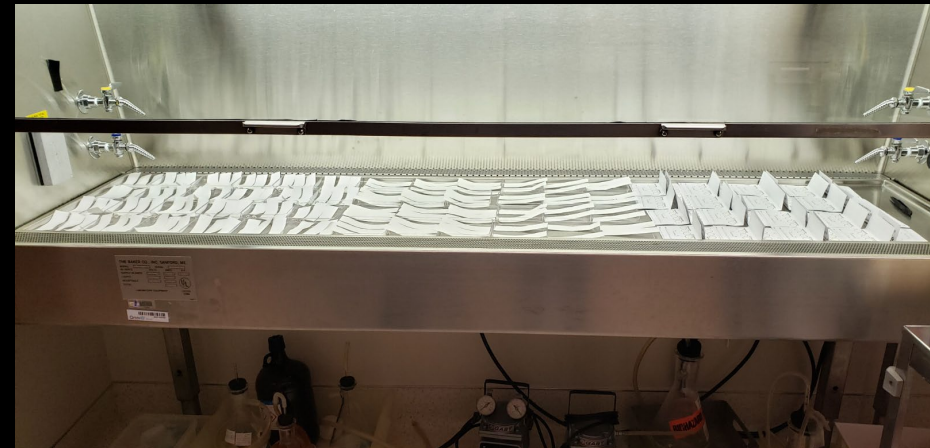
Dry Technology: Validation

Long-term Stability and Quantifying Loss

Verified:

1. Best paper type for the majority of analytes
2. Best storage conditions Vs -80C
3. Duration of stability for each analyte
4. Rehydration and extraction methods

Still actively working to improve the recovery for Additional analytes.



Liquid Collections

Phlebotomy and Passive Saliva Samples

- “Liquid” bio samples are collected pre and post to establish a baseline immune profile.
- Will act as a control for the dry bio sample collections.



ARTEMIS II Sampling Matrix

Pre-Flight Samples

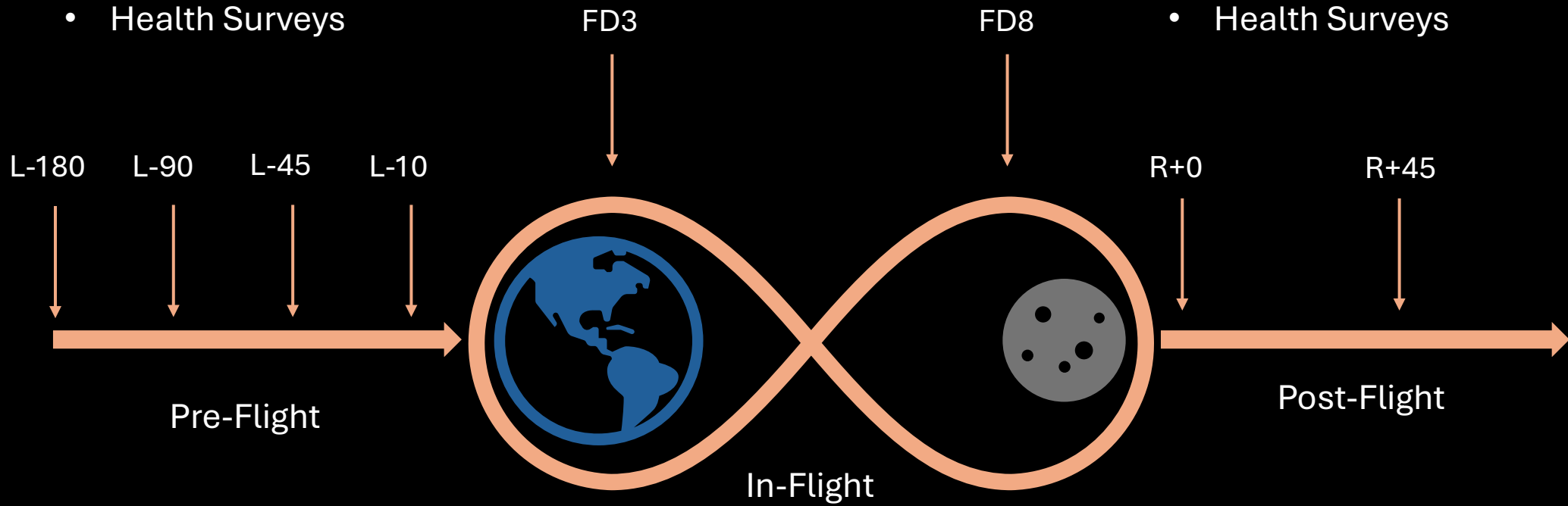
- Viral Detection Book
- Biomarkers Book
- Dry Blood Card
- Liquid Saliva
- Liquid Blood
- Health Surveys

In-Flight Samples

- Viral Detection Book
- Biomarkers Book
- Dry Blood Card (Art III)

Post-Flight Samples

- Viral Detection Book
- Biomarkers Book
- Dry Blood Card
- Liquid Saliva
- Liquid Blood
- Health Surveys



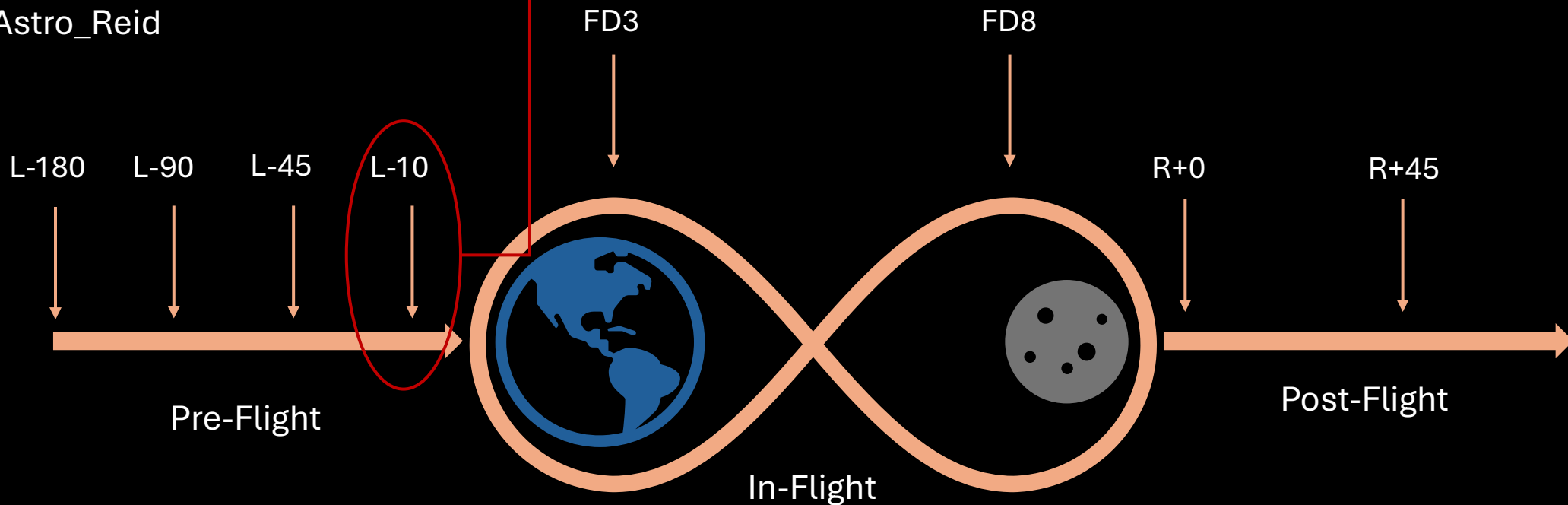
Current Study Status



Astro_Reid

As of March -

- Crew trained on how to properly perform dry collections
- Liquid blood collection processed for each timepoint
 - Plasma isolations/WBC Phenotyping/T-Cell Function Assays
- Awaiting April launch
 - Repeating L-10s due to mission slips
- Updating and improving dry technology



Deep Space Countermeasures

Pre-Flight Countermeasures:

- Clinical History Screening
- Vaccinations and Medications

In-Mission Countermeasures:

- Functional food enriched diets
- Probiotics
- Vitamin D
- Aerobic/Resistive exercise
- Stress relieving exercises/VR meditation
- Medications (Anti-Histamines, Valacyclovir, etc.)
- Monitoring
 - Virus PCR, WBC differential, soluble measures



Countermeasures-based Improvements in Stress, Immune System Dysregulation and Latent Herpesvirus Reactivation onboard the International Space Station – Relevance for Deep Space Missions and Terrestrial Medicine



Brian E. Crucian^{a,*}, George Makedonas^b, Clarence F. Sams^a, Duane L. Pierson^a, Richard Simpson^c, Raymond P. Stowe^d, Scott M. Smith^a, Sara R. Zwart^e, Stephanie S. Krieger^f, Bridgette Rooney^g, Grace Douglas^a, Meghan Downs^a, Mayra Nelman-Gonzalez^f, Thomas J. Williams^a, Satish Mehta^b

Exercise as a countermeasure for latent viral reactivation during long duration space flight

Nadia H. Agha¹ | Satish K. Mehta² | Bridgette V. Rooney^{1,3} | Mitzi S. Laughlin¹ |
Melissa M. Markofski¹ | Duane L. Pierson⁴ | Emmanuel Katsanis^{7,8} | Brian E. Crucian⁴ |
Richard J. Simpson^{1,5,6,7,8}

Specific Immunologic Countermeasure Protocol for Deep-Space Exploration Missions

George Makedonas^{1†}, Satish Mehta^{1†}, Alexander Choukèr^{2†}, Richard J. Simpson^{3,4,5}, Gailen Marshall⁶, Jordan S. Orange⁷, Serena Aunon-Chancellor⁸, Scott M. Smith⁹, Sara R. Zwart⁹, Raymond P. Stowe¹⁰, Martina Heer¹¹, Sergey Ponomarev¹², Alexandra Whitmire¹³, Jean P. Fripiat¹⁴, Grace L. Douglas⁹, Stephanie S. Krieger¹³, Heman Lorenzi¹⁵, Judith-Irina Buchheim², Geoffrey S. Ginsburg¹⁶, C. Mark Ott⁹, Meghan Downs¹³, Duane Pierson⁹, Natalie Baecker¹¹, Clarence Sams⁹ and Brian Crucian^{9*†}

The Immunology and Virology Laboratory Team !!!

