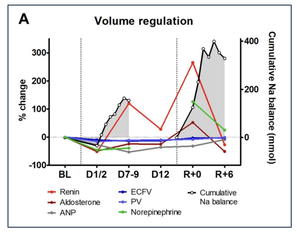
Transcriptomic Mechanisms of Microgravity-Induced Renal Sodium Retention

Ayma Rizwan - Terre Haute South Vigo High School, Terre Haute, IN.

Introduction

- As the 2nd most metabolically active organ, the kidney is central to understanding organisms' adaptations to extreme environments such as spaceflight.
- Cephalad fluid shift contributes to spaceflight-related conditions including **SANS**, **Orthostatic intolerance** after re-entry, **Reduced renal perfusion** (blood flow to the kidneys).[1]
- Over prolonged exposure to micro(μ)gravity, plasma volume decreases, and the body reduces signals that normally promote sodium excretion.

Figure 1: Long duration spaceflight → increased sodium reabsorption → increased cumulative sodium balance.[2]

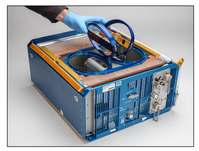


Metadata Analysis

Spaceflight conditions

- NASA OSDR OSD-513[3] collected as part of RR-23 payload by SpX-21 mission
- Male mice (C57BL/6J wild type), ~16-17 weeks in age (Kidneys at this age are fully mature but not yet affected by aging-related decline)
- Launched into space (n=10) or maintained as ground controls (n=10) under identical conditions.

Figure 2: Rodent Flight Hardware, as used on the OSD-513[3] spaceflight group and control group.



- Rodent Flight Hardware with 12h light/12h dark (to mimic standard circadian cycle)
- Ad libitum food schedule with Nutrient Upgraded Rodent Food Bars.

RNA collection and sequencing

- 38 days in space → mice returned to Earth alive → euthanized by bilateral thoracotomy with sedation → kidney tissue collected.
- Post-tissue collection: each left kidney immersed in RNALater for 24 hours at 4˚C → RNA extracted and stored at -80 ˚C → left kidneys sequenced.
- Raw FASTQ files run through quality control, trimmed, and aligned to the reference genome prior to quantification of gene counts.

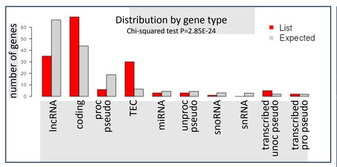


Figure 3: DEG show significant enrichment for protein-coding transcripts ($p = 2.85 \times 10^{-24}$), supporting the biological relevance of pathway-level findings and indicating that results are not driven by nonfunctional or predicted genes.[4]

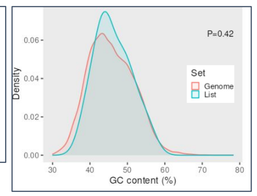


Figure 4: Genes are distributed across chromosomes with no significant clustering ($p = 0.2$), indicating no locus-specific bias in expression changes.[4]

Differential Gene Expression (DEG) Analysis

- Jupyter notebook DGEA (Python's implementation of DESeq2)
- PCA and Volcano plots (to visualize differential gene expression)
- ShinyGo's KEGG analysis, DAVID, STRING (to evaluate enriched pathways and identify protein function)

Confirmed Mechanisms

- PPAR signaling (Ces1g, Lrat, Nr4a3, Hmgcs2, Apoa1, Fabp1, Cyp4a14)
- Altered metabolic/mitochondrial capacity, indicating reduced active transport of Na^+ (Cox6a2, Hmgcs2, Mat1a, Eif4ebp3, Bhmt)

Transcriptomics Analysis & Results

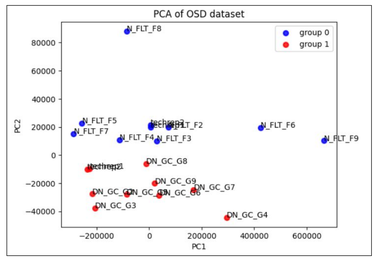


Figure 5: PCA Plot depicting distinctive expression patterns between the spaceflight (in blue) and ground control (in red), proving μgravity effect on mice gene expression is statistically strong enough to separate samples by condition.

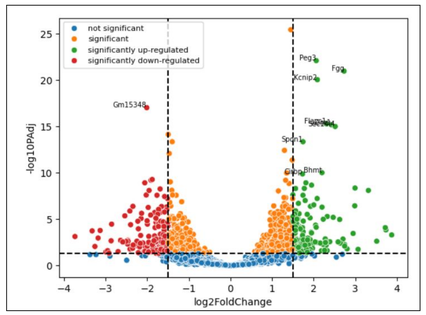


Figure 6: Volcano plot prioritizing genes for pathway enrichment and biological interpretation.

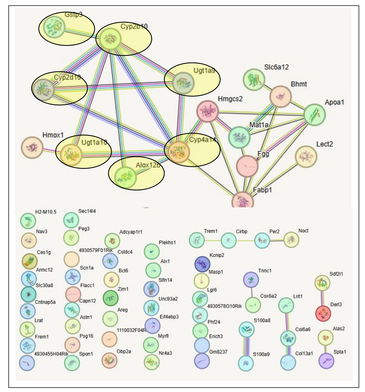


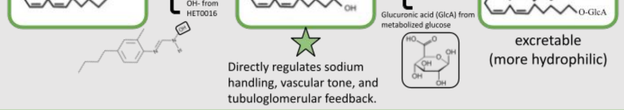
Figure 7: Nodal network of proteins representing interactions between products of DEG.[5]

Predicted Interactions	Others	Known Interactions
gene neighborhood	textmining	from curated databases
gene fusions	co-expression	experimentally determined
gene co-occurrence	protein homology	

Figure 8: Selected Upregulated Genes' Relevance [5][6]

Family	Relevance	Genes
Cytochrome pathway:	• Accelerated production of 20-HETE.	Cyp4a14, Cyp2b10, Cyp2d10
UDP-glucuronosyltransferase pathway:	• Glucuronidation enzymes→ makes molecules more excretable. • Prevents buildup of AA metabolites.	Ugt1a9, Jgt1a10
Gstp3 (Glutathione S-transferase pi 3)	Neutralizes reactive oxygen made during AA oxidation & eicosanoid synthesis.	Gstp3
Arachidonate 12-lipoxygenase, 12R-type	Oxygenates AA, producing 12(R)-HpETE, indirectly reducing 20-HETE production.	Alox12b

Figure 9: Simplification of Cytochrome P450 (CYP450) arachidonic acid metabolism pathway[7] (by Ayma).



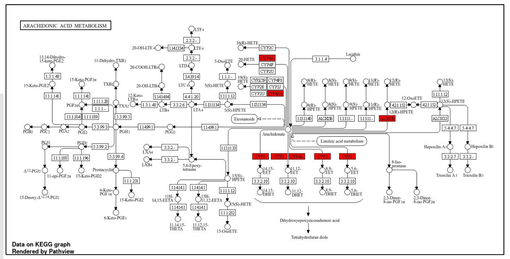
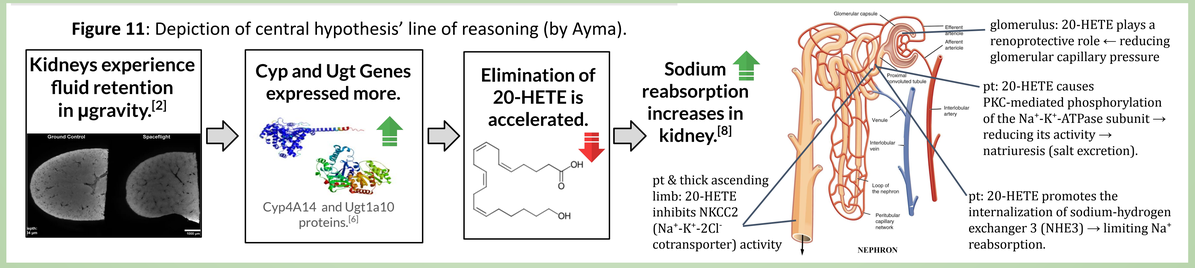
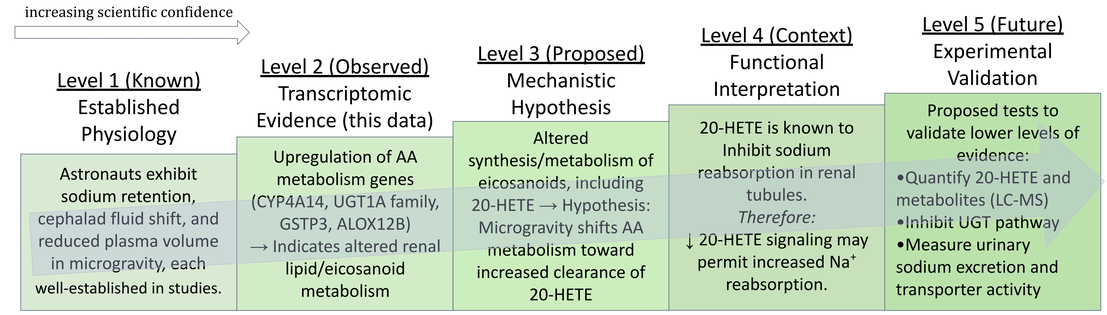


Figure 10: Full AA Metabolism Pathway, generated by QuickGo's KEGG, with the dataset's DEG in red, contextualizing gene-level changes within the broader metabolic pathway.[4]

Figure 11: Depiction of central hypothesis' line of reasoning (by Ayma).



Evidence Ladder



Proposed Experiments

Is altered 20-HETE metabolism a mechanism of sodium retention in microgravity by way of upregulation of Cyp and Ugt genes (1), and can it be modulated pharmacologically (2)?

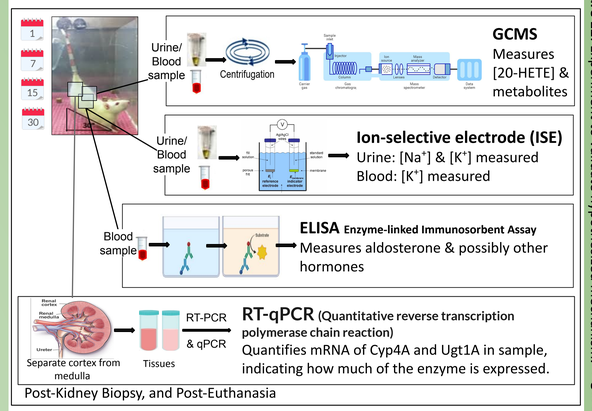
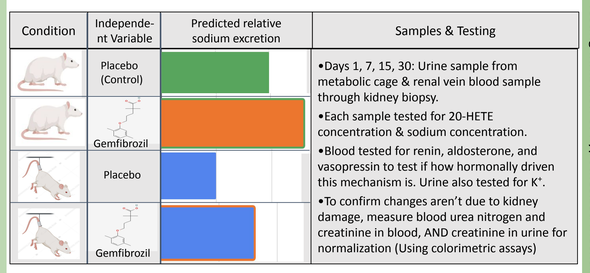


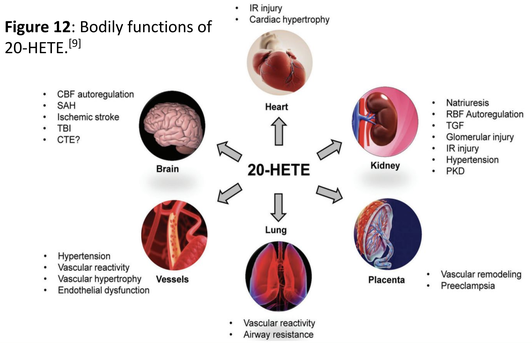
Figure 12: Experiment to validate hypothesized mechanism. **Figure 13:** Testing inhibition of hypothesized mechanism.



Significance & Future Direction

- Development of a novel, aldosterone-independent mechanism of fluid regulation mediated by local renal lipid signaling.
- Future work will experimentally test & identify potential therapeutic targets for spaceflight-induced fluid shifts and related disorders.

Figure 12: Bodily functions of 20-HETE.[9]



References Cited

[1] Norsk, P., Asmar, A., Damgaard, M., & Christensen, N. J. (2015). Fluid shifts, vasodilatation and ambulatory blood pressure reduction during long duration spaceflight.

[2] Olde Engberink, R.H.G., van Oosten, P.J., Weber, T. et al., npj Microgravity 9, 29 (2023). The kidney, volume homeostasis and osmoregulation in space: current perspective and knowledge gaps.

[3] Galazka , J., Zawieja , D., Gebre, S., Costes , S., Boyko , V., Dinh , M., Chen, Y., Houseman , C., Lai Polo, S., Peach , K., Vallota-Eastman, A., Saravia-Butler, A., Oribello , J., Torres , A., Novak , B., Yehia , J., & Han, C. (2025). Transcriptional profiling of kidneys from mice flown on the RR-23 mission. NASA Open Science Data Repository, Version 5. https://osdr.nasa.gov/bio/repo/data/studies/OSD-513

[4] QuickGO. (2001, August). Www.ebi.ac.uk. https://www.ebi.ac.uk/QuickGO/annotations

[5] STRING. (2000). STRING: functional protein association networks. String-Db.org. https://string-db.org/

[6] SMART. (1998, May). Smart.embl.de. https://smart.embl.de/

[7] Tallima, H., & El Ridi, R. (2018). Arachidonic acid: Physiological roles and potential health benefits – A review. Journal of Advanced Research, 11, 33–41. https://doi.org/10.1016/j.jare.2017.11.004

[8] Gross Anatomy of the Kidney | Anatomy. (2026). Lumenlearning.com. https://courses.lumenlearning.com/suny-dutchess-anatomy-physiology/chapter/gross-anatomy-of-the-kidney/

[9] Fan, F., & Ye, Y. (2016, June 1). Molecular mechanisms and cell signaling of 20-hydroxyeicosatetraenoic acid in vascular pathophysiology. Semantic Scholar. semanticscholar.org/paper/Molecular-mechanisms-and-cell-signaling-of-acid-in-Fan-Ge/a3359ba8eaea9271403014541a1ee8124c39af04