Characterizing SERCA function in murine skeletal muscles after 35 days of spaceflight

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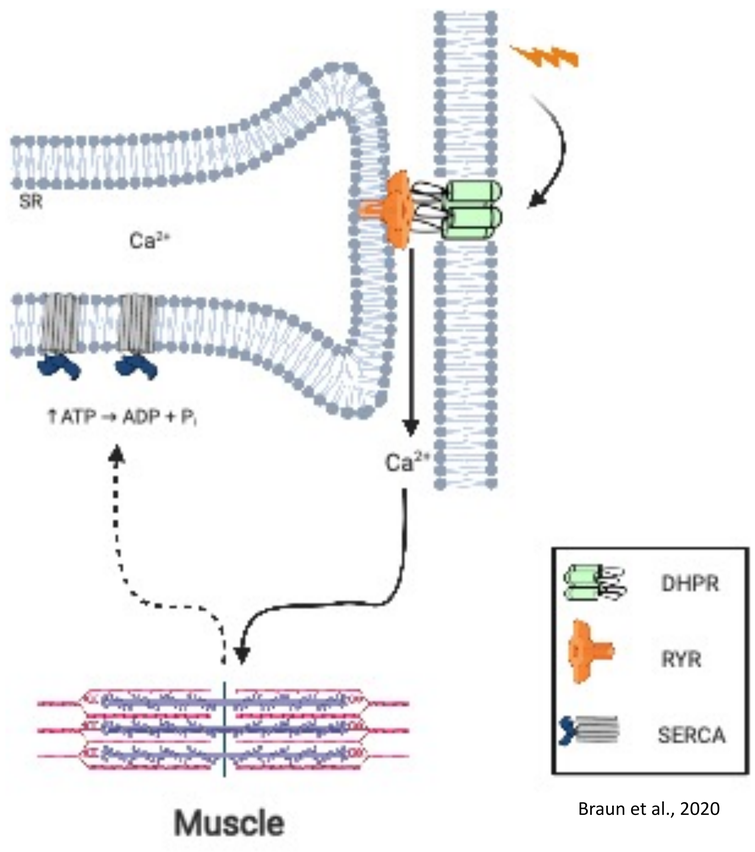
Introduction

1. Spending time in a microgravity environment is known to lead to muscle atrophy and weakness[1]
2. SERCA regulates $[Ca^{2+}]_i$ by catalyzing the active transport of Ca^{2+} into the sarcoplasmic reticulum and is critical for healthy muscle function[2].
3. SERCA dysfunction can lead to increased $[Ca^{2+}]_i$ resulting in cellular damage and death, contributing to muscle atrophy and weakness
4. **The potential role of SERCA dysfunction in post-spaceflight muscle atrophy and weakness has yet to be investigated and remains unknown.**



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Purpose

The purpose of the study was to characterize SERCA function, its regulators, and reactive oxygen/nitrogen species (RONS) in the soleus and tibialis anterior (TA) muscles of male and female mice after ~35-37 days in space.

Methods

- **C57BL/6 Mice**
  - Mice were launched at 10 weeks of age and spent between 35 and 37 days housed on the International Space Station
  - Two control groups were included in the study:
    - VIV: housed in standard rodent cages
    - GC: housed in the same cages as the flight group
- **Ca^{2+} Uptake Assay**
  - Measured using a Molecular Devices M2 plate reader and Indo-1 fluorescent indicator
- **Western blotting**
  - Western blots were performed to assess SERCA2a, SERCA1a, PLN, SLN, and NNAT protein content as well as the presence of RONS

Results

Table 1. Spaceflight induces a significant loss in muscle mass. TA muscle weights are significantly lower in the flight group compared to the two control groups (*$p < 0.05$). The RR9 mission (males) and RR1 mission (females) similarly show significant reductions in muscle weight post-flight (**$p < 0.01$; ***$p < 0.005$). (n = 4 per group, RR1; n = 9-10 per group, RR9)

	VIV	GC	Flight
RR9 *Tibialis Anterior*	53.4 ± 5.0*	53.0 ± 3.4*	47.3 ± 5.7
RR9 *Soleus*	8.2 ± 1.2***	7.9 ± 0.9**	6.1 ± 0.7
RR1 *Soleus*	7.0 ± 0.5**	7.0 ± 0.4**	5.0 ± 0.8

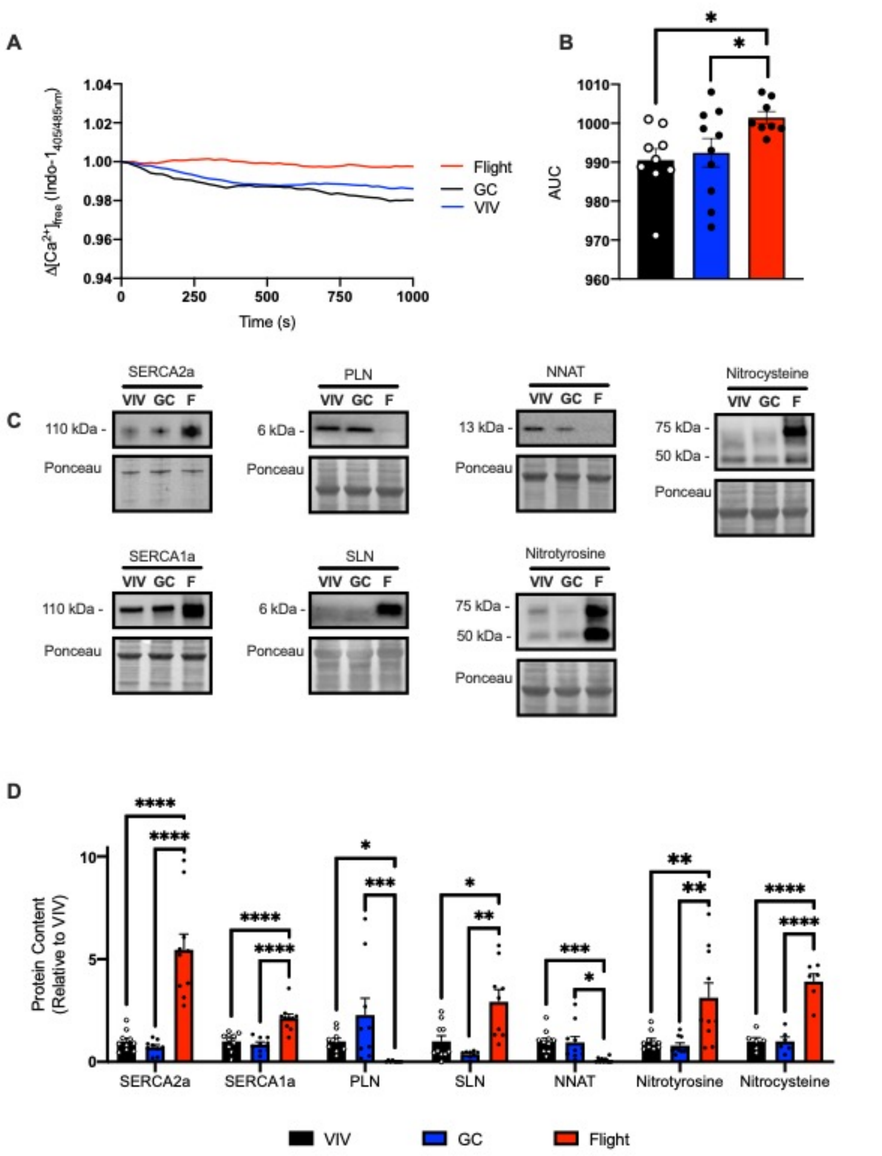

Figure 1. Spaceflight increases SERCA Ca^{2+} uptake, alters SERCA regulatory protein content and increases RONS in the soleus muscle of male mice. Ca^{2+} uptake (A) is significantly reduced in the flight group, represented by increased AUC (B). Western blotting was employed to investigate SERCA2a/1a, PLN, SLN, and NNAT protein content as well as the presence of RONS (C,D). *$p < 0.05$; **$p < 0.01$; ***$p < 0.005$; ****$p < 0.0001$ (n = 9-10 per group).

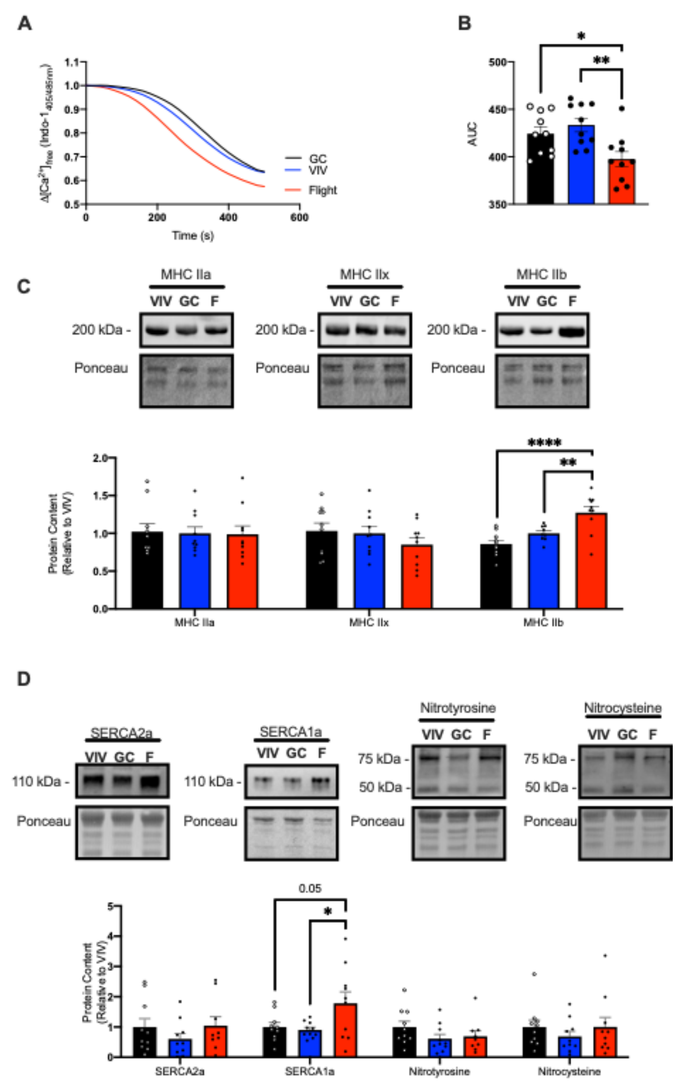

Figure 2. Spaceflight increases SERCA Ca^{2+} uptake and induces a shift towards a fast phenotype in the TA of male mice. Ca^{2+} uptake (A) is significantly increased in the flight group, represented by decreased AUC (B). Western blotting was employed to investigate MHC IIa, IIx, and IIb (C), SERCA2a/1a protein content as well as the presence of RONS (D). *$p < 0.05$; **$p < 0.01$; ****$p < 0.0001$; values above bars indicate p values (n = 9-10 per group).

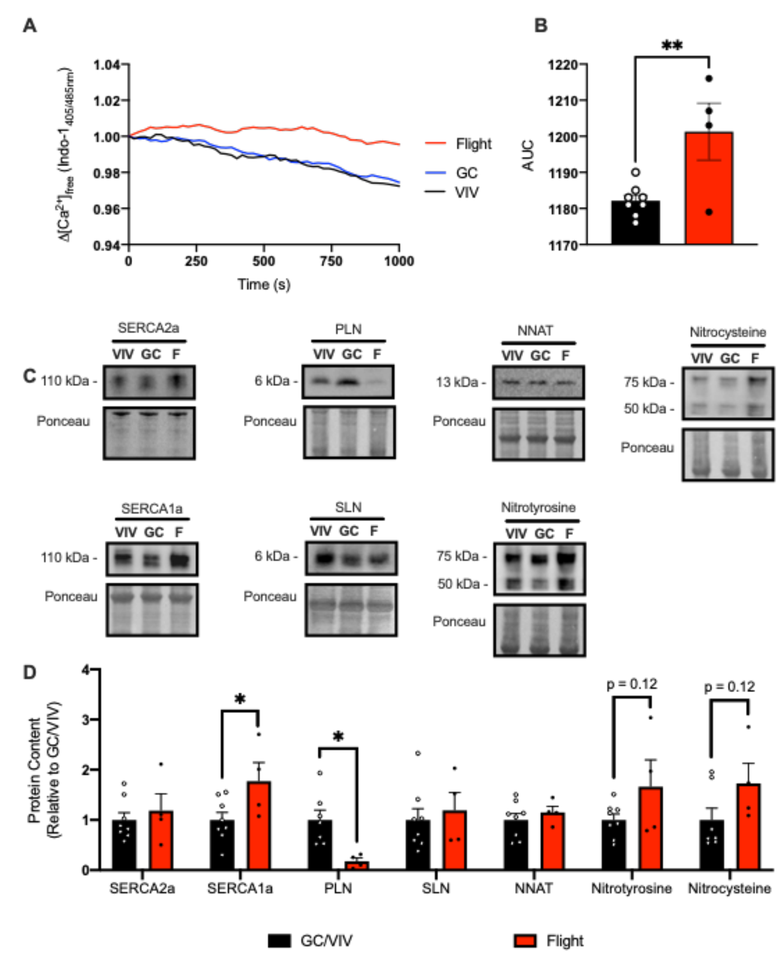

Figure 3. Spaceflight reduces SERCA Ca^{2+} uptake, alters SERCA regulatory protein content and increases RONS in the soleus muscle of female mice. Ca^{2+} uptake (A) is significantly reduced in the flight group, represented by increased AUC (B). Western blotting was employed to investigate SERCA2a/1a, PLN, SLN, and NNAT protein content as well as the presence of RONS (C,D). Black bars indicate combined GC/VIV control groups. *$p < 0.05$; *$p < 0.01$; **$p < 0.01$; values above bars indicate p values (n = 4-8 per group).

Conclusions

- **Spaceflight induces SERCA dysregulation and impaired calcium uptake in the postural soleus of male and female mice**
  - SERCA content is increased with dynamic changes in SERCA regulator content
  - Impairment in SERCA calcium uptake may be due to increased RONS
- **Spaceflight induces a fiber type shift towards a fast-glycolytic muscle and improves calcium uptake in the TA**

References & Acknowledgements

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