MicroAge Mission: Examining the Effects of Microgravity and Electrical Stimulation on the Proteome of Human Tissue-Engineered Muscle Constructs.

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Shifting age demographics are contributing to increasing numbers of older adults with poor health. The basis of age-related muscle loss has yet to be fully uncovered, however we have previously demonstrated that skeletal muscle from aged humans and mice show attenuated responses to exercise. This is particularly apparent in redox-regulated responses to contractile activity. In an analogous yet accelerated manner, the muscles of astronauts exposed to microgravity (μg) also rapidly lose mass and are relatively unresponsive to training in spaceflight.

As part of a UK Space Agency funded mission to the International Space Station (ISS), we developed tissue-engineered, human muscle constructs to delineate proteomic changes in μg and on earth with and without a period of electrical stimulation. Muscle constructs were integrated into bioreactors designed to autonomously perform electrical field stimulations and monitor muscle contractions via impedance spectroscopy. Upon return from the ISS, samples underwent LC-MS using a timsTOF mass spectrometer in dia-PASAF mode. Bioinformatic analyses determined differential expression patterns and gene ontology (GO) term functional enrichment.

In total, 2934 human proteins were identified across all samples. When examining the effects of μg on the proteome at rest, 541 proteins were differentially expressed (DE) (287 upregulated and 254 downregulated, $p < 0.05$) compared with ground samples. Using GO analysis, enriched pathways included nucleotide metabolic processes, mitochondrial gene expression and oxidative metabolism. Electrical stimulation of muscle constructs on ground resulted in 357 DE proteins versus rest, whilst in μg there were 272. Of these, 57 were shared between μg and ground. Electrical stimulation on ground resulted in the enrichment of muscle system processes, muscle contraction pathways and protein folding. However, these processes were perturbed in μg, top GO terms were associated with wound healing, muscle processes and ribonucleoprotein biogenesis. Further, in depth analyses of redox-regulated pathways are on-going.