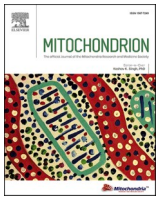
Mitochondrion 76 (2024) 101855

Contents lists available at ScienceDirect

Mitochondrion

journal homepage: www.elsevier.com/locate/mito



Review

Mitochondrial stress in the spaceflight environment

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ARTICLE INFO

Keywords:
Mitochondria
Spaceflight
Radiation
Microgravity
Oxidative stress

ABSTRACT

Space is a challenging environment that deregulates individual homeostasis. The main external hazards associated with spaceflight include ionizing space radiation, microgravity, isolation and confinement, distance from Earth, and hostile environment. Characterizing the biological responses to spaceflight environment is essential to validate the health risks, and to develop effective protection strategies. Mitochondria energetics is a key mechanism underpinning many physiological, ecological and evolutionary processes. Moreover, mitochondrial stress can be considered one of the fundamental features of space travel. So, we attempt to synthesize key information regarding the extensive effects of spaceflight on mitochondria. In summary, mitochondria are affected by all of the five main hazards of spaceflight at multiple levels, including their morphology, respiratory function, protein, and genetics, in various tissues and organ systems. We emphasize that investigating mitochondrial biology in spaceflight conditions should become the central focus of research on the impacts of spaceflight on human health, as this approach will help resolve numerous challenges of space health and combat several health disorders associated with mitochondrial dysfunction.

1. Introduction

While Space agencies prepare for future missions to the Moon, and eventually to Mars and beyond, our understanding of the health risks associated with space travel remains incomplete. Conditions experienced by humans in space and even the controlled conditions of spacecraft are hostile. Space travel upends the performance of almost every biological system. The exact conditions that individuals are subjected to depend on the duration and the path of travel, thus, individuals will experience different conditions on a several days-long flight to the Moon, and a several months-long space travel to Mars which would include an extended period of exploration. NASA plans to move beyond International Space Station (ISS) residing at low-Earth-orbit (LEO) and land astronauts on the Moon by 2025, to next establish a permanent presence on the Moon in preparation for sending humans to Mars within Artemis Program. Moreover, with the Commercial Crew Program we see greater involvement of the private sector in space exploration. Companies like Axiom, Northrop, StarLab, and Orbital Reef aim to develop commercial space modules, SpaceX, Boeing, BioServe, and Space Tango are leading in technological solutions, and Virgin Galactic, BlueOrigin aim at space tourism. Several of these companies already succeeded in bringing passengers to space, and SpaceX and Axiom resupply and organize crewed missions to the ISS. However, the longest current space missions to the ISS lasted over a year, and longer missions beyond LEO involve additional challenges. Therefore, providing a safe travel experience for an average passenger, rather than only highly trained selected astronauts, must be a priority in space health (Table 1).

Space remains a challenging environment that deregulates an individual's homeostasis (Simonsen et al., 2020). The main external hazards associated with spaceflight include space radiation, microgravity, isolation/confinement, distance from Earth, and hostile environment (Afshinnekoo et al., 2020). The primary risks to human health and performance arise from these five hazards, but space radiation, microgravity, and isolation play the most significant roles in causing cellular damage. Additionally, factors such as physiological conditions and genetics can also impact individual responses to these extreme conditions.

Characterizing the biological responses to the spaceflight environment is essential to validate the risks, and to develop effective protection strategies. Organisms have evolved an array of biochemical and physiological responses that allow them to maintain internal homeostasis under challenging environments. In recent years, it has been demonstrated that the conditions of spaceflight can induce cellular stress, including stress to mitochondria (Silveira et al., 2020; McDonald et al., 2020; Luxton et al., 2020; Singh et al., 2021; Nguyen et al., 2021;

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https://doi.org/10.1016/j.mito.2024.101855
Received 8 July 2023; Received in revised form 12 February 2024; Accepted 22 February 2024
Available online 23 February 2024

Table 1
Table of Abbreviations.

Abbreviation	Definition
NASA	The National Aeronautics and Space Administration, of the USA
ISS	International Space Station
LEO	Low Earth Orbit
GCRs	Galactic Cosmic Rays
SPEs	Solar Particle Events
HZE	High atomic number (Z) and energy (E) particles
EMF	Electromagnetic fields
ELF-EMF	Extremely low-frequency electromagnetic fields
Sv, mSv	Sievert, milliSievert, the unit of absorbed radiation dose by weight
Gy, mGy	Gray, milliGray, the unit of absorbed ionizing radiation dose
Hz, MHz, GHz	Hertz, Megahertz, Gigahertz, the unit of frequency
T, μT	Tesla, microTesla, the unit of magnetic field strength
W, mW	Watt, milliWatt, the unit of power
SMG	Simulated Microgravity
G	Gravitational acceleration constant, 9.81 m/s^2
mtDNA	Mitochondrial DNA
nDNA	Nuclear DNA
ATP	Adenosine triphosphate
ADP	Adenosine diphosphate
ETS	Mitochondrial electron transport system
OXPHOS	Mitochondrial oxidative phosphorylation
ROS	Reactive oxygen species
TCA cycle	Tricarboxylic acid cycle, Krebs cycle
NADPH	Nicotinamide adenine dinucleotide phosphate
SOD	Superoxide dismutase, enzyme
GPx	Glutathione peroxidase, enzyme

Michaletti et al., 2017; Afshinnekoo et al., 2020). Indeed, mitochondrial stress is considered one of the fundamental features of space travel (Silveira et al., 2020; Afshinnekoo et al., 2020), and a focal point for growing number or researchers, including NASA GeneLab Analysis Working Group (Silveira et al., 2020). Therefore, in this paper, we summarize how the hazards of spaceflight impact mitochondrial function (Fig. 1). Because mitochondria lay at the very center of the spaceflight phenotype and many physiological processes, and are affected by all of the main spaceflight hazards, we emphasize that investigating the effects of spaceflight on mitochondria should be put in the focus of more ground and flight-based biology studies.

2. Main hazards of spaceflight

2.1. Space radiation

Space radiation consists of ionizing radiation from the sun (solar particle events, SPEs), and the galaxy (galactic cosmic rays, GCRs). On Earth, organisms have evolved under levels of radiation filtered through our planet's atmosphere and magnetosphere, protecting us from harmful cosmic radiation. The majority of the solar wind particles are at low energies and pose little risk, but the solar wind can become dangerous in case of sudden outbursts of energy called SPEs, consisting of ionizing radiation, i.e. high-frequency electromagnetic fields (EMF) including Gamma rays (with frequencies about 10^{20} Hz and above) and X rays (about 10^{17}–10^{19} Hz), which can vary in composition and intensity according to the size of the SPE (Reitz et al., 2012). However, the most threatening type of ionizing radiation in space is GCRs, with high frequencies of about 10^{23} Hz. GCRs consist of protons and heavy ions, which include about 87 % protons, 12 % ions, and 1 % so-called HZE particles (high atomic number and energy). HZE are nuclei stripped of their orbital electrons that have travelled through the galaxy before entering our solar system moving nearly at the speed of light (Reitz et al., 2012).

The Earth's magnetosphere capture a majority of SPEs and GCRs in radiation belts (Reitz et al., 2012), so the natural background radiation on Earth is about 2.5 mSv/year (milliSievert/year), but can be about 5 mSv/year at high altitudes. The Moon, however, has no atmosphere or magnetic field, and therefore no protective radiation belt, so the space radiation reach the surface of the Moon at doses that can exceed 1 Sv/year (Sievert/year). Moreover, in interplanetary space the exposure to space radiation can be even higher, about 700 times higher than on Earth (Kim et al., 2021). SPEs and GCRs are highly ionizing forms of radiation that can penetrate biological tissues (Simonsen et al., 2020). Without countermeasures, the SPEs and GCRs can ionize cells and cause damage to macromolecules including DNA, in consequence increasing oxidative stress and DNA mutations, and decreasing cellular repair. So, this types of space radiation pose an especially challenging health risk for upcoming missions to the Moon and Mars. On Earth, radiation

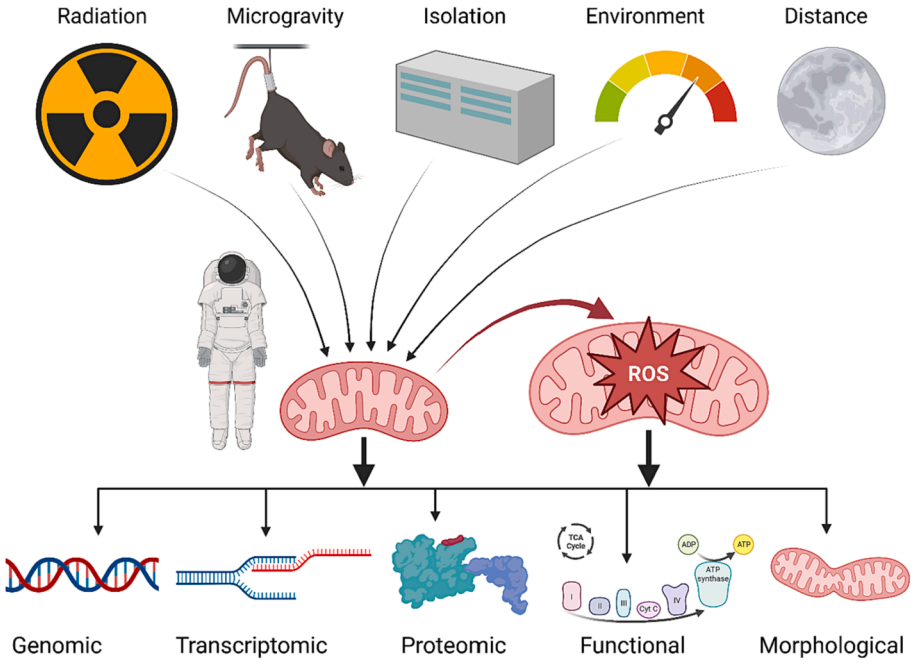

Fig. 1. The main health hazards of space flight: radiation, microgravity, isolation and confinement, challenging environment, and distance from Earth, cause direct or ROS-induced changes to mitochondria at genomic, transcriptomic, proteomic, functional and morphological levels in various tissues of the astronauts and in ground-based studies.

exposure can be simulated by applying known doses of X and gamma rays (Zhang et al., 2018; Liu et al., 2013), and GCRs can be simulated with a simplified five-ion protocol, that consists of protons H^1 and heavy ions Si^{28}, He^4, O^{16}, Fe^{56}, normalized to 500 mGy (milliGray), developed at NASA Space Radiation Laboratory (Simonsen et al., 2020).

Other types of radiation associated with space travel include non-ionizing, low-frequency EMF. UV light is also a type of non-ionizing radiation present in space, but the astronauts are usually well protected from the UV with space suits, helmets, and shielding of the spacecraft. Further, the non-ionizing radiation includes microwaves (10^{10}–10^{12} Hz), and radio waves (10^7-10^9 Hz), but the organisms are also exposed to extremely low-frequency electric fields (10^3 Hz and below) and magnetic fields from electrical wiring (0.1 μT). Moreover, all organisms on Earth and astronauts within LEO are also exposed to the magnetic field from the Earth's magnetosphere (25–65 μT).

2.2. Microgravity

Organisms evolved in the presence of the Earth's gravitational field of 1 G (gravitational acceleration of 9.81 m/s^2). Microgravity is experienced when the gravitational field is lower than that on Earth. On Mars, microgravity is 0.376 G, and on the Moon only 0.166 G, and can be even lower, or close to zero in interplanetary space. Microgravity can cause numerous effects on organisms, including body fluid redistribution. During spaceflight, the bodily fluids shift from the lower limbs toward the head and upper parts of the body (Nelson et al., 2014). This process mildly changes the placement of some organs and modifies the function of the cardiovascular system. In addition, microgravity causes major muscle atrophy, bone mass loss, osteoporosis, and kidney stones (Nelson et al., 2014). However, the muscle and bone dystrophies can be to some extent reduced by regular exercise. In ground-based studies microgravity can be simulated using several simulated microgravity (SMG) protocols, including bed rest, clinostats, random positioning devices, centrifuges, or hind limb unweighting.

2.3. Isolation/confinement, distance from Earth, hostile environment

More stressors related to space travel are induced by isolation in a relatively small space, without access to normal levels of physical activity and human interaction. Lack of activity is to some extent compensated by an onboard exercise routine. However, confinement with several co-inhabitants changes the sensory environment, which can induce stress (Raber et al., 2021; Hollis et al., 2022), and it prohibits normal social interactions and dynamics within the group. Additional stress is caused by distance from Earth, imposing communication delays between the ship and Earth, which makes it substantially more difficult to assist the astronauts in emergencies. So, distance from Earth requires implementation of an onboard medical care system, which may prove insufficient in serious medical emergencies. Moreover, following the best effort, the hostile environment of a closed spacecraft remains challenging to space travellers. On board a spacecraft, the temperature, humidity, O_2 and CO_2 concentration, noise, and light regime are regulated by the life-supporting systems which aim to simulate conditions on Earth. But for example, the disruption of the light–dark cycle poses an environmental stressor that cannot be underestimated, as light regulates several hormonal functions (Walton et al., 2011). Importantly, the individual responses to spaceflight conditions are modified by several individual factors like physiological conditions, genetics, age, and sex.

3. Mitochondrial stress in animal physiology

Mitochondria are heterogeneous organelles performing a variety of complex functions within the cells, which include cellular energy and heat production, maintaining Ca^{2+} homeostasis and membrane potential, Fe/S cluster synthesis, lipid synthesis and oxidation, amino-acid, ascorbate, and one-carbon metabolism, neurotransmitters synthesis and degradation, ROS production, steroidogenesis, intracellular and apoptotic signalling (Monzel et al., 2023). Mitochondria are also capable of dynamically and reversibly adapting to energetic and environmental conditions, including external stressors, often challenging the simplistic 'healthy function' vs 'dysfunction' paradigm (Monzel et al., 2023).

In the classic understanding, mitochondria are responsible for energy synthesis in the form of adenosine triphosphate (ATP) via oxidative phosphorylation (OXPHOS) in mitochondria through the electron transport system (ETS). Mitochondrial DNA (mtDNA) contains 13 genes which encode subunits of enzyme complexes of the ETS and the ATP synthase, whereas some mitochondrial genes are also encoded by nuclear DNA (nDNA). During OXPHOS, the electrons harvested from food-derived substrates flow through protein complexes of the ETS to fuel the passing of protons from the mitochondrial matrix into the intermembrane space, and protons passing back through the ATP synthase power the phosphorylation of ADP to ATP (Koch et al., 2021). The rate of OXPHOS fluctuates with changes in substrate availability and ATP demands. Thus, the efficiency of mitochondrial respiration is a determinant of the energetic capacity of the tissues, and the whole organism. Electrons escaping the ETS can react with oxygen to form reactive oxygen species (ROS), which contribute to oxidative stress. When exposure to ROS exceeds the quenching capacity of antioxidants, the cells experience oxidative stress, which contributes to oxidative damage to macromolecules, such as lipids, proteins, and DNA (Finkel and Holbrook, 2000). ROS have been considered inevitable by-products of OXPHOS, with the majority of ROS production occurring within the mitochondria (Bratic and Trifunovic, 2010). However, ROS also act as signaling molecules that mediate cellular and physiological processes (Zuo et al., 2015), and mild oxidative stress can even increase mitochondrial function by upregulating antioxidants, repair enzymes and mitochondrial biogenesis (Hood et al., 2018).

The efficiency of mitochondrial respiration ultimately controls the amount of energy uptake from the environment and the relative allocation of this energy to support processes throughout the body (Koch et al., 2021). Therefore, mitochondria energetics is the key mechanism underpinning many ecological, physiological, and evolutionary processes (Koch et al., 2021). This includes variations in physiological performance and life histories, such as whole-organism metabolic rate (Salin et al., 2016), adaptation to environmental conditions (Treberg et al., 2018; Sokolova, 2018), growth (Barneche et al., 2019), reproductive success (Hood et al., 2018), and accelerated ageing (Speakman et al., 2004). Moreover, the capacity to deal with mitochondrial stress varies among species (Treberg et al., 2018) and individuals (Salin et al., 2015).

4. Mitochondrial stress in space flight

Numerous experiments in space, mainly on the ISS revealed the detrimental effects of spaceflight on mitochondria. Mitochondrial stress can be considered a central driver of the biological impact of space travel (Silveira et al., 2020; Afshinnekoo et al., 2020). An integrated NASA GeneLab analysis using several space missions, multiple tissues, and cell lines revealed that spaceflight impacts mitochondrial function at the genetic, protein, and metabolite levels (Silveira et al., 2020), which has also been supported in other studies (Garrett-Bakelman et al., 2019; Chakraborty et al., 2020; Luxton et al., 2020; Vernice et al., 2020).

Mitochondria changes at functional and genomic levels have been identified in post-flight human and mouse twins versus ground-based controls, where on top of the modified nDNA and mtDNA OXPHOS genes expression, the post-flight twins had reduced antioxidant defenses, and increased markers of oxidative stress 8OHdG and PDGF2-alpha in urine (Silveira et al., 2020; Garrett-Bakelman et al., 2019). In the Twin Study, following a year-long astronaut mission to the ISS (Garrett-Bakelman et al., 2019), increased inflight mtRNA in blood plasma was correlated with the time spent in the orbit. Treating muscle cells with plasma from the astronaut twin showed increased basal ADP-

induced respiration, and decreased reserve ATP-linked capacity, but no change in maximum respiration, which was consistent with the changes in the mtRNA levels. In addition, increased lactic acid, and the lactic/pyruvic acid ratio, were observed in the urine and plasma of the astronaut twin, indicating a shift from aerobic towards anaerobic metabolism, and an increased TCA cycle was observed in plasma. Spaceflight also altered the astronaut gene expression in pathways associated with aerobic metabolism, mitochondrial transport and apoptosis, and hypoxia (Garrett-Bakelman et al., 2019). Moreover, analysis of human cell lines after the ISS flight showed changes in the expression of overlapping genes across all four cell types i.e. fibroblasts, endothelial cells, primary T cells, and hair follicle cells, which contained multiple mitochondrial function genes, associated with mitochondrial ATP synthesis, electron transport system, OXPHOS, and proton transportation (Silveira et al., 2020).

Transcriptomics and proteomics analysis of data from Rodent Research 1 and 3 missions, including various tissues like liver, kidney, eye, and adrenal gland from two mouse strains C57BL/6 and BALB/c housed on the ISS for about 30 days, showed that cellular stress was linked to mitochondrial, ribosomal and translational changes (Silveira et al., 2020). The ribosome assembly and mitochondrial translation pathways were altered in most of the tissues, implying disrupted mitochondrial protein homeostasis. In addition, the data showed contrasting changes in the nDNA and mtDNA expression of mitochondrial genes, for example, downregulation of nDNA-coded OXPHOS transcript in the liver, including cytochrome *c* oxidase, but upregulation of mtDNA-coded OXPHOS transcripts like mt-ND4 and mt-ND5. Similar changes were found in kidneys, and partially in muscles, suggesting that spaceflight suppresses nDNA-coded OXPHOS gene expression and that the over-stimulation of the mtDNA genes may act as a compensation mechanism towards the diminished mitochondrial metabolism (Silveira et al., 2020). The transcriptional data revealed also changes in several metabolic pathways, for example, key citric acid cycle enzyme - citrate synthase decreased in muscle but increased in the liver, and malate dehydrogenase, carnitine acetyltransferase, and malonyl-CoA decarboxylase decreased in the muscle (Silveira et al., 2020). Data from gastrocnemius and quadriceps muscles of Rodent Research 9 mice showed increases in several mitochondrial-related pathways, such as β-oxidation of long-chain fatty acids, tricarboxylic acid cycle, L-carnitine and malate (Silveira et al., 2020). The quadriceps muscle of C57 mice that were flown to the ISS in Rodent Research 4 showed reduced muscle weight, reduced mtDNA copy numbers and metabolomic changes (Chakraborty et al., 2020). Genes promoting muscle atrophy were upregulated, whereas genes encoding proteins linked to muscular functions were down-regulated. The network linked to mitochondrial function showed differentially expressed genes and metabolites, including those associated with synthesis and hydrolysis of ATP, generation of reactive oxygen species, mitochondrial permeability, calcium usage, and inhibited tricarboxylic acid cycle, potentially driving energy-deprived state (Chakraborty et al., 2020).

In more studies, three astronauts experienced elevated mitochondrial mtDNA and mtRNA in their blood and urine, indicating persistent mitochondrial stress, chronic oxidative stress, inflammation, and rapid telomere shortening after return to Earth from long-term six months to one-year flights to the ISS (Luxton et al., 2020). Alterations of protein content and function of the mitochondria in cardiac tissue, which consists of mononuclear cells with a high density of mitochondria, caused by spaceflight are associated with cardiac atrophy. The presence of mitochondrial DNA and RNA in the peripheral blood of the astronauts means that these molecules are being lost due to cardiac atrophy (Vernice et al., 2020). Moreover, oxidative stress associated with space flight induces damage to the retina in human eyes, including ageing-like and diabetes-like degeneration, such as cataracts and an increase in intraocular pressure (Nguyen et al., 2021). Mitochondrial oxidative damage to lipids and DNA in ocular tissue and increased apoptosis of retinal and photoreceptor cells have also been reported in mice after space shuttle flight compared to ground-based controls. Moreover, the expression of mitochondria-associated apoptotic genes and genes associated with oxidative stress were upregulated (Mao et al., 2012). Lastly, in-flight and post-flight astronaut hair follicle samples after 6 months on the ISS, had reduced mtDNA to nDNA ratios, decreased expression of several redox-related genes, and increased expression of genes related to oxidative stress (Indo et al., 2016).

Astronauts in space are usually exposed to several health hazards at the same time, and because the main hazards usually act simultaneously, their effects are difficult to separate experimentally. These effects cannot be separated while on space missions, but they can be separated in experiments using a simulated ground-based approach, such as mentioned before simplified five-ion protocol for GCRs, clinostat protocols for simulated microgravity, or analog-astronaut protocols for isolation and confinement.

5. Radiation

5.1. Ionizing radiation: galactic cosmic rays (GCRs), gamma rays, X-rays

The major hazard of space exploration is ionizing radiation, especially cosmic radiation including GCRs (McDonald et al., 2020). A combination of highly ionizing protons and heavy ions with a chronic dose rate is characteristic of GCRs exposure in space, and can lead to genomic and functional damage to the mitochondria. During future trips to Mars, the astronauts will be exposed to about 350 mSv/year of ionizing radiation, consisting mainly of GCRs, but also of gamma and X-rays (Afshinnekoo et al., 2020). Mitochondria are a common target of ionizing radiation because as a numerous organelle, they occupy large areas of the cells (Afshinnekoo et al., 2020). Mitochondrial gene expression is susceptible to ionizing radiation, and dysregulation in the expression of certain gene families is consistent with the dysregulation of mitochondrial respiratory function (McDonald et al., 2020).

A combined ground-based and spaceflight database revealed a trend of increased activation of multiple mitochondrial-related pathways as a consequence of ionizing radiation (McDonald et al., 2020). Several biological pathways became dysregulated after cosmic radiation exposure including those associated with mitochondrial function, oxidative stress, and ER protein assembly (McDonald et al., 2020). Mitochondrial ROS production induced by cosmic radiation leads to dysregulated expression of nDNA and mtDNA-coded genes associated with OXPHOS (Afshinnekoo et al., 2020). In addition to changes in gene expression, radiation-induced mitochondrial ROS can cause damage to mtDNA, nDNA, and proteins, including the OXPHOS enzymatic complexes, and lipids. Moreover, oxidative damage to mtDNA tends to be several-fold higher than in nDNA, due to the absence of protective histones and DNA repair mechanisms (Afshinnekoo et al., 2020). An imbalance between the expression of nDNA and mtDNA-coded genes may lead to compensatory expression of some proteins, and thus to disrupted mitochondrial protein proportions and assembly, which promote the mitochondrial unfolded protein response (mtUPR). High levels of ionizing radiation-induced oxidative stress also contribute to metabolic disorders and accelerated ageing, and even to cancer, which is one of the main risks of long-term spaceflight (Afshinnekoo et al., 2020).

In ground-based studies, simulated GCRs protocol has been used in several experiments with rodents revealing consequences to nervous and musculoskeletal system mitochondria (Simmons et al., 2022; Kim et al., 2021). Balb/c mice subjected to 50 cGy of simulated GCRs had impaired short-term and spatial memory, and differential expression of over a hundred proteins, including those associated with mitochondria function, like cytochrome *c*, mitochondrial complex I, and NADH dehydrogenase (Simmons et al., 2022). In rodents, GCRs can also induce depression and cognitive impairments (Raber et al., 2021). Retina from the eyes of rats exposed to 1 and 4 Gy proton beams was investigated for mitochondria-associated genes and protein expression profiles. The expression of several genes involved in the regulation of the

mitochondrial apoptotic pathway was up-regulated after radiation, including Bbc3, Bax, Bak1, Bid, and Bcl2, and genes involved in oxidative stress, including Sod2, Gpx, and Ucp3, whereas Ucp2 was down-regulated, and the apoptotic cells after irradiation showed greater cytochrome *c*-release (Mao et al., 2012). Moreover, cardiomyocytes from C57BL/6NT mice exposed to 90 cGy (900 mSv) of protons and iron ions for 28 days showed major differentially expressed genes associated with cell cycle and oxidative responses. The top pathways with changed expression including ones involved in oxidative phosphorylation and mitochondrial function, but also those associated with Parkinson's, Alzheimer's, Huntington's diseases, cardiac muscle contraction, and cardiomyopathy, which share many energy-related transcripts (Coleman et al., 2015). Osteoclast cells from Balb/c mice three months following exposure to 0.5 Gy (500 mSv) of total body simulated GCR, showed increased levels of mitochondrial reactive oxygen species without changes to mitochondrial mass. Also, the basal, ATP-linked and proton leak mitochondrial respiration and non-mitochondrial respiration were increased in these osteoclasts, whereas utilization of some substrates like pyruvic, citric, succinic acid, and fumaric acid was decreased, indicating metabolic rewiring of the mitochondria (Kim et al., 2021).

Other types of ionizing radiation in space include Gamma rays and X-rays, both of which have been shown to affect mitochondria functions. Mice from C57BL/6 strain and Nox2 knock-out mice deficient the NADPH oxidase, which is a source of brain ROS, placed on Cobalt57 plates emitting a low dose gamma radiation of 0.04 Gy for 21 days had their brains isolated for analysis of oxidative stress markers (Mao et al., 2017). Oxidative stress to lipids and activity of anti-oxidative enzyme superoxide dismutase (SOD), was increased in the C57 mice one month after the exposure, whereas the Nox2 deficient mice were partially protected (Mao et al., 2017), indicating the role of endogenous ROS in coping with the radiation. Mitochondria of human keratinocyte epithelial cells and hamster ovarian fibroblast cells irradiated with 5 mGy, 0.5 Gy or 5 Gy gamma rays demonstrated loss of OXPHOS enzymes function already four hours post-irradiation which was dose-dependent, followed by recovery in some complexes twelve to ninety-six hours later. The cells also showed irregular mitochondrial protein synthesis, mtDNA damage, and increased mitochondrial genome frequency (Nugent et al., 2010). Whereas, radio-resistant *Caenorhabditis elegans* exposed to chronic gamma-ray irradiation suffered increased ROS production and dose-rate-dependent activation of SOD1 and GPx antioxidant enzymes (Maremonti et al., 2020). At dose rates above 40 mGy the formation of free radicals caused oxidative stress-induced transcriptomic response, including changes in mitochondrial function, protein degradation, dysregulation of genes involved in DNA damage and repair, and chromosome segregation. Moreover, the irradiation lead to a reduced number of offspring per individual, accompanied by down-regulation of over 300 genes associated with the reproductive system, gamete development and apoptosis (Maremonti et al., 2020).

Mice exposed to doses of ionizing X radiation of 0.2 and 2 Gy, had their cardiac mitochondria isolated for analysis of functional alterations and proteomics. The proteomics showed radiation-induced deregulation of twenty-five proteins, mainly associated with oxidative phosphorylation, pyruvate metabolism, and cytoskeletal structure. Moreover, the mitochondria showed partial deactivation of Complex I and III, decreased succinate-induced respiratory capacity, increased ROS, and oxidation of mitochondrial proteins (Barjaktarovic et al., 2011). Radiation-induced lung injury in dogs was associated with ROS production, increased MnSOD, increased copy numbers of mitochondrial genes, and an initial decrease followed by an increase in mitochondrial complexes activity (Yin et al., 2019). X-rays are also the cause of radiation-induced heart disease after treatment with targeted irradiation of breast or lung cancers, which has been shown to have profound effects on cardiac mitochondria (Livingston et al., 2020). Livingston et al., 2020 discuss more than thirteen works describing the impact of therapeutic doses of radiation that lead to radiation-induced heart disease, which was associated with morphologic, functional, genomic, transcriptional, and apoptotic changes of cardiac mitochondria in humans and rodents (Livingston et al., 2020).

5.2. Non-ionizing, low-frequency radiation

In the modern world, humans and many animals are surrounded by low-frequency EMF. This non-ionizing radiation includes microwaves from mobile phones and WIFI, radio waves and TV broadcasts. Moreover, organisms are often exposed to extremely low-frequency electromagnetic fields (ELF-EMF) from power lines and electrical wiring, as well as magnetic fields generated by the electrical wiring, and are always under the magnetic field of the Earth. The astronauts in space are also surrounded by these types of fields, which are an integral part of the space environment and an integral part of the automatic systems of the spacecraft. Although not much is known about the impact of these fields on mitochondrial function, most of these fields likely do not cause substantial damage to mitochondria.

A potential health hazard associated with exposure to microwaves has long been under speculation. Indeed, some studies suggest that microwaves at 2.1 GHz, and 0.120 mW/cm^2, which is within the wireless exposure limits in the US and Canada, can cause a mild mitochondrial disorder, such as hyperpolarization of mitochondria demonstrated in increased membrane potential and cytochrome *c* levels, and cell apoptosis (Esmekaya et al., 2017). In another study, hypoxia-inducible factor-1α (HIF-1α) was upregulated in neuron-like cultured cells, which in turn initiated a protective mechanism by activating the expression of cytochrome *c* oxidase subunits COXIV, particularly COXIV-2 and COXIV-1 (Hao et al., 2018).

Radio waves have been studied in cancer therapy for tumours with poor response to conventional treatments, and suggested to be non-destructive to normal cells (Curley et al., 2014). Mitochondrial morphology, membrane potential, mitochondrial oxygen consumption, and ROS production were investigated in human pancreatic cancer cells. Radiofrequency of 13.6 MHz altered the morphology and polarisation of the mitochondrial membrane, impaired mitochondrial respiration, and increased ROS production, and a mitochondrial marker Tom20 was found in phagosomes, indicating the elimination of damaged mitochondria (Curley et al., 2014). Similar potential cancer treatment was evaluated in human cancer cell lines after exposure to ELF-EMF associated with electric currents at 50 Hz, and resulted in reduced cell proliferation, increased mitochondrial enzymatic activity and membrane potential, without changes in ATP production, changed mitochondrial protein profile, downregulated expression of phospho-ERK, p53 and cytochrome *c*, but no transcriptional changes of respiratory complexes (Destefanis et al., 2015).

5.3. Mitochondrial hormesis

Interestingly, the negative effects of radiation on mitochondria are not always persistently harmful, and in some cases can improve performance. Our group has shown that in house mice (Zhang et al., 2018) the negative effect of X-ray exposure on mitochondria can be short-lived, with acute X-ray exposure (5 Gy) inducing an initial drop in mitochondrial respiratory function, an increase of ROS production and oxidative damage, followed by an upregulation of mitochondrial complex function. Within ten days, each mitochondrial respiratory capacity, ROS levels, and oxidative damage all returned to baseline (Zhang et al., 2018). Our recent data showed decreased respiratory capacity in liver and skeletal muscle mitochondria of deer mice exposed to 2 Gy X-ray irradiation, which returned to baseline in the muscle after a month of recovery (unpublished data). Further, a study in copepods showed that UV light increased the proportion of mitochondrial junctions and density of the inner mitochondrial membrane, but decreased mitochondrial size, and induced early-life reproductive output, while reducing longevity (Heine et al., 2021). The observed recovery of mitochondrial

performance in mice and increase in the reproductive performance of copepods represent mitohormetic responses, where radiation-induced ROS stimulates the upregulation of cellular defense mechanisms including antioxidants, and rates of mitochondria turnover and repair (Zhang et al., 2018; Heine et al., 2021).

6. Microgravity

Microgravity is the main environmental hazard in space travel. Microgravity disrupts cellular homeostasis, and as a consequence leads to oxidative stress, DNA damage, genetic mutations, and decreased cellular viability. Mitochondrial oxidative stress has been described after exposure to real and simulated microgravity in various tissues, including nervous, musculoskeletal and cardiovascular systems (Nguyen et al., 2021).

In the hippocampus, a brain region involved in learning, memory, and navigation, mitochondria undergo gene expression changes as a consequence of microgravity (Rubinstein et al., 2021; Ji et al., 2022; Wang et al., 2016). Mice subjected to SMG by hindlimb unloading for 30 days in single or paired housing had dysregulated cytokines associated with inflammation in the hippocampus (Rubinstein et al., 2021). After 7 and 21 days of SMG by tail suspension, rats developed changes in the expression of 67 and 42 mitochondrial metabolic genes in the hippocampus respectively, as well as downregulated complexes I, II, IV, and malate dehydrogenase, and upregulated expression of mitochondrial anti-oxidative stress proteins DJ-1 and peroxiredoxin 6, but no obvious elevated cellular apoptosis (Wang et al., 2016). Rats subjected to microgravity by tail suspension for 28 days had elevated mitochondria number and size in the hippocampus, and 163 differentially expressed proteins, with 128 upregulated and 35 downregulated, altering several mitochondrial processes including the TCA cycle and fatty acids oxidation (Ji et al., 2022), which was suggested to be associated with a decline in cognitive function. Mitochondria disruption after microgravity can also induce oxidative damage in cerebral arteries (Zhang et al., 2014). A significant increase in mitochondrial ROS levels, mitochondrial permeability transition pore opening, and markers of lipid peroxidation, and a decrease in mitochondrial membrane potential, respiratory exchange ratio, and antioxidant enzyme MnSOD/GPx activity have been observed in cerebral arteries of rats exposed to SMG by four-week hind limb unloading (Zhang et al., 2014). Whereas, treatment with NADPH oxidase inhibitor and mitochondria-targeted antioxidant promoted recovery of the mitochondrial function, regulated via mitochondrial expression of NADPH oxidases.

Cardiac and skeletal muscles of rats after just six days of exposure to microgravity showed increased expression of malate dehydrogenase, and the levels of mRNAs encoding subunits of COX, without changes in its activity in the cardiac muscle, which was however decreased in skeletal muscle (Connor and Hood, 1998). Human osteoblasts exposed to microgravity had decreased mitochondrial protein levels, stimulated glycolysis, and disrupted Krebs cycle at succinate to fumarate transformation. In addition, several mitochondrial complexes were deregulated, with downregulated complex II and IV by 50 % and 14 % respectively, upregulated complex III by 60 %, and with reduced coupled ATP synthesis, and decreased antioxidant enzymes (Michaletti et al., 2017). These effects of microgravity on osteoblasts' mitochondria were dominant, and authors speculated that a decrease in ATP levels associated with the down-regulation of CoQ10 could justify the chronic fatigue experienced by astronauts (Michaletti et al., 2017). Moreover, in human endothelial cells after 4 and 10 days of SMG in rotating wall vessel a marker protein of mitophagy was increased, and mitochondrial content and maximal respiratory capacity were reduced, suggesting phenotypic changes in response to meeting the metabolic challenges caused by SMG (Locatelli et al., 2020). Cultured human and insect lymphocytes after microgravity associated with space flight and SMG on clinostat show cytoskeletal and morphological alterations resulting in mitochondrial clustering towards one side of the cells, disrupted mitochondrial transport along microtubules, altered mitochondrial cristae morphology, and increased number of apoptotic cells (Schatten et al., 2001). Furthermore, simulated microgravity was associated with changes in several mitochondria-related characteristics in *C.elegans*, including activation of mitochondrial UPR, mitochondrial ROS production, and mitophagy, but reduction of mitochondrial oxygen consumption and mitochondrial membrane potential, followed by decreased locomotion behaviour (Liu et al., 2019).

Interestingly, the apoptotic effects of microgravity also impact several types of cancer cells and therefore, microgravity may have anticancer effects through its effects on cellular growth inhibition. Some studies even suggest it as a novel therapeutic approach different from conventional chemo and radiotherapy (Jeong et al., 2018). SMG is associated with reduced mitochondrial activity and elevated autophagy and apoptosis in glioma (Takeda et al., 2009), lymphoma (Jeong et al., 2018), leukemic cells (Singh et al., 2021), and breast cancer (Zheng et al., 2012). Tumour growth rate and mitochondrial activity were also reduced, and sensitivity to the anti-cancer agent cisplatin increased in glioma cells exposed to SMG (Takeda et al., 2009). The Hodgkin's lymphoma cells decreased their mitochondrial mass, ATP levels, ATPase and ATP synthase, increased their ROS production and NADPH oxidase expression and underwent autophagy via activation of AMPK and MAPK pathways, and inhibition of mTOR (Jeong et al., 2018). The leukemic cells had decreased proliferation and viability, enhanced ROS formation, and increased DNA damage and expression of DNA damage sensing proteins, which led to apoptotic cell death (Singh et al., 2021). Rotary cell culture of human breast cancer cells DA-MB-231 developed oxidative stress and DNA damage after just 48 h of SMG, however, a different cell line MCF-7 increased its mitochondrial activity and demonstrated minimal oxidative damage. The activity of superoxide dismutase increased in the MCF-7 cells and decreased in MDA-MB-231 cells. SMG promoted the expression of ERα and PKC and decreased the activity of MDA-MB-23 the ERα negative cells, but not MCF-7 the ERα positive cells (Zheng et al., 2012), so the anticancer effects of microgravity are not universal and depend on the type of cancer and cancer cell lines.

7. Isolation and confinement, distance from Earth, and hostile environments

Isolation and confinement on Earth can cause chronic stress that alters individual physiology, including elevated cortisol levels, and elevated immune responses, it also has profound effects on the central nervous system, including behavioural changes and cognitive impairments, upregulated oxidative stress pathways, and inflammatory responses (Tahimic et al., 2019; Rubinstein et al., 2021). Although astronauts are not in complete solitude, they operate in the confined and isolated environment of a spacecraft during both the short and long space flights. Moreover, a company of the same crew members for a long time can induce social stress.

Recent evidence suggests that social stress may induce mitochondria dysfunction, neuroinflammation, and even stress-related depression (Hollis et al., 2022). Several types of induced psycho-social stress in rodents decreased mitochondrial energy production capacity which was manifested in reduced respiratory complexes activity, rate of mitochondrial respiration, mitochondrial membrane potential, mitochondrial content, and mtDNA copy number, accompanied by changes in mitochondrial shape and size (Picard and McEwen, 2018). Furthermore, isolation can modify immune responses to spaceflight in combination with other stressors such as simulated microgravity. Hindlimb unloading in individually housed mice led to a reduction in circulating immune cells, which was not observed in socially housed mice (Tahimic et al., 2019). MCAT mice, which overexpress human catalase, an enzyme responsible for anti-oxidative response, display decreased oxidative damage and mitochondrial ROS production (Rubinstein et al., 2021). The wild-type mice subjected to 30 days of hindlimb unloading showed dysregulated cytokines associated with hippocampus inflammation, and

the highest expression was found in mice housed individually in opposition to ones housed socially, whereas the MCAT mice have been protected from these adverse changes in cytokine expression in both housed individually and socially (Rubinstein et al., 2021). This indicates that the effect of isolation can have serious consequences for mitochondria-mediated processes in space travel.

The hostile environment of spaceflight involves exposure to regulated temperature, carbon dioxide levels, disturbed circadian rhythms, stress, and constant noise on board the spaceship where the crew must spend extended time periods. As temperature and O_2 levels are usually maintained at stable levels, the CO_2 may be elevated, leading to mild hypoxia. Hypoxia is regulated by a family of transcription factors, named the hypoxia-inducible factors (HIFs) subunits HIF-1α and HIF-2α that are degraded in an oxygen-dependent manner (Thomas and Ashcroft, 2019). The HIFs signalling supports anaerobic ATP production by downregulating OXPHOS, thus reducing the oxygen-dependent energy production and causing metabolic reprogramming under hypoxia. Hypoxia can dysregulate the mitochondrial TCA cycle, downregulate OXPHOS, and stimulate ROS production, but the HIFs signalling is capable of mitigating the oxidative damage by upregulating SOD expression, synthesis of glutathione, and mitochondria turnover (Thomas and Ashcroft, 2019).

Disturbed circadian rhythms also disturb mitochondrial functions, because circadian sleep-wake cycling interacts with redox, bioenergetics, and temperature regulation. It has been suggested, that nuclear DNA repair and protein synthesis are maximized during the wake phase, whereas the mitochondrial remodelling is maximized during the sleep phase, indicating that the wake phase is more protective and restorative to the nucleus, whereas the sleep phase is more protective and restorative to the mitochondria (Richardson and Mailloux, 2023). Mitochondria are a major cellular source of ROS which contributes to oxidative damage, and in humans, enzymatic antioxidants like catalase, GPx, and SOD tend to peak during the light phase, whereas non-enzymatic antioxidants like melatonin, and vitamin C and E tend to peak during the dark phase. Moreover, the light–dark cycle regulates mitochondrial anion carrier proteins including uncoupling proteins (UCPs) and adenine nucleotide translocase (ANT). ANT catalyses the exchange of ADP anion for ATP synthesis, and peroxidation products travel through the mitochondrial membrane via UCPs and ANT, stimulating the proton leak. Modifying the mitochondrial coupling efficiency determines the balance between proton leak associated with heat production, and oxidative phosphorylation associated with ATP production, which also regulates ROS production. In mice, the activity of UCP2 linked to glucose-induced ATP production has been upregulated in the resting phase, and mitochondrial fatty-acid β-oxidation is also promoted to be greatest during the sleep phase. Moreover, anion carriers such as UCPs are regulated by several mediators including ROS, GSH, thyroid hormones, and melatonin. So, the function of melatonin includes the regulation of sleep and clock proteins, but also protection against mitochondrial ROS by dissipating the protein gradient. Melatonin, which is mainly synthesized in mitochondria, inhibits mitochondria fission and is excreted in the dark or sleep phase, allowing fission during the wake phase (Richardson and Mailloux, 2023). Therefore, the effects of disturbed woke-sleep cycles on mitochondrial function in spaceflight should not be underestimated.

Lastly, a great distance from Earth can cause stress, including acute, and chronic unpredictable stress, which is of significance for mitochondrial function (Picard and McEwen, 2018), and the stress hormone cortisol can also play a role in lowering mitochondrial oxygen consumption (Richardson and Mailloux, 2023). Moreover, serious health issues in space may be hard to address without access to adequate medical attention and difficulties in obtaining timely help while being so far from home.

8. Interactions between the main hazards of spaceflight

On Earth, organisms have evolved within the context of strong gravitational pull and low levels of radiation, however in space, these proportions are reversed. Importantly, the main hazardous factors of spaceflight such as space radiation, microgravity, and isolation are difficult to separate, as they are usually all present during the space flight at the same time. These hazards have been shown to interact and thus, the impact on organisms' physiology is different than the sum of their independent impacts. In many cases, the observed effects would be higher than the additive, thus defined as synergistic.

Microgravity has been shown to interact with space radiation to influence organism health at a cellular level. A variety of experiments investigated if the effects of microgravity are manifested independently of space radiation (Yatagai et al., 2019). For instance, the level of 4-HNE protein, a marker for lipid peroxidation, was elevated in mice brain cortex and hippocampus after hindlimb unloading and irradiation, but the expression of SOD was decreased in the mice exposed to either only microgravity or combined microgravity and radiation (Mao et al., 2016). Moreover, an additive effect on double-strand breaks, a type of complex DNA damage, has been observed in a variety of cell cultures, but not on DNA repair. However ground-based studies indicate that hindlimb unloading and whole-body irradiation in mice skeletal muscle, decrease levels of endothelial nitric oxide synthase, and SOD, but increase levels of xanthine oxidase (Yatagai et al., 2019). These examples indicate that the synergistic effects of radiation and microgravity can modify the response to oxidative stress that is associated with mitochondrial ROS production. Moreover, the interaction of microgravity with isolation can change the way that both factors affect mitochondria-mediated processes. Social in contrast to individual housing of mice subjected to simulated microgravity lead to elevated circulating CD4 T immune cells, and differentially expressed inflammatory cytokines in the hippocampus of both wild-type and MCAT mice (Tahimic et al., 2019; Rubinstein et al., 2021). The mice housed individually under simulated microgravity suffered the most adverse effects.

Importantly, oxidative damage accumulation depends also on the physiological condition of the organism, which can be further induced by external hazards such as space radiation or microgravity in space, which can also lead to synergistic effects. For instance, wound healing is associated with the process of oxidative shielding of mitochondria i.e. change of mitochondrial function profile from meeting normal metabolic needs, to pro-inflammatory mitochondria producing oxylipin and ROS for anti-inflammatory defence, and releasing mtRNA to activate inflammasomes and interferon (Naviaux, 2019). During this process, the OXPHOS mitochondrial respiration drops, and energy production shifts towards glycolysis, so cellular aerobic metabolism is compromised, but it shifts back to OXPHOS when the wound recovery occurs (Naviaux, 2019). A synergistic effect of combined exposure to SMG, ionizing radiation, and cortisol has been shown in cultured human fibroblasts, demonstrating that these spaceflight hazards interfered with wound healing (Radstake et al., 2023). Cortisol exposure reduced expression of pro-inflammatory cytokine interleukin 1-alpha, as well as cortisol exposure in combination with radiation or SMG. But exposure to SMG or radiation in combination with SMG without cortisol increased the interleukin expression. Cortisol also modified the expression of interleukin 6, cytokines, and several growth factors associated with different stages of wound healing (Radstake et al., 2023).

9. Individual factors modifying susceptibility to spaceflight hazards

The degree to which intracellular stress responses and recovery are being induced by a spaceflight varies with the lifestyle and genetic predispositions of the individual.

Physical exercise is considered one of the countermeasures against the detrimental effects of the main hazards of spaceflight (Kim et al.,

2021). Overall, physical exercise has been shown to reduce the risk of cardiovascular, endocrine, musculoskeletal, immunological, and oncological diseases, and repeated exercise induces protective cellular adaptations via improving mitochondrial biogenesis, OXPHOS capacity, and resilience to oxidative stress (Powers et al., 2016). Mitochondria in skeletal muscle tissue can undergo rapid changes as a result of exercise, for instance, endurance training can increase mitochondrial volume by up to 50 % within a few weeks, and cause a shift of substrate metabolism toward higher reliance on lipids (Michaletti et al., 2017). Physical exercise is used to counter alterations to cardiovascular and musculoskeletal systems, including cardiovascular deconditioning, muscular atrophy and bone loss, after prolonged exposure to microgravity in spaceflight, so astronauts are advised to exercise about 2 h every day (Gao et al., 2023). Systematic research based on data from three space agencies revealed that physical exercise decreased space radiation-induced DNA damage, oxidative stress, and inflammation, and increased antioxidant activity (Kim et al., 2021). While exercise alone cannot completely counter the negative effects of spaceflight (Silveira et al., 2020), the effects sizes were high for the pre-exposure, and the highest for the post-exposure physical training in rodents, and in humans also during the exposure (Kim et al., 2021). Moreover, exercise improves vascular endothelial function and mitigates arterial stiffening, reduces inflammation and oxidative damage, and increases antioxidant enzymes. Mitochondria play a central role in exercise-stimulated vascular health benefits, as exercise induces mitochondrial adaptations through the increasing release of ROS and NO that activate multiple intracellular signalling pathways and alleviate mitochondrial fragmentation (Gao et al., 2023).

Although mitochondrial stress is a consistent phenotype of space travel, the ability to maintain mitochondrial homeostasis under any stressor can differ among individuals (Treberg et al., 2018). For example, gene regulation varies among different strains of lab mice subjected to spaceflight, with C57BL/6 altering the translation of more genes than BALB/c mice (Silveira et al., 2020). Further, about 25 % of astronauts on the ISS develop ocular problems, such as spaceflight-associated neuroocular syndrome (SANS), which appears to be linked to specific genetic traits that disrupt the one-carbon-metabolism pathway, and is associated with oxidative stress (Schmidt and Goodwin, 2013), and appears to be more prominent in males. Moreover, the phenotypic differences among mice and humans are less pronounced on Earth than in space suggesting that the phenotypes attributable to genotypic differences among individuals can be amplified in the extreme environment of space (Schmidt and Goodwin, 2013). Considerable differences are also found between species of the same taxa. For example, a white-footed mouse after exposure to gamma (90–417 cGy) radiation had higher mortality than a laboratory mouse (Liu et al., 2013), but despite their prolonged lifespan, the white-footed mice developed more adrenal and liver tumors compared to lab mice, and anemia following the irradiation. The authors indicated that the different response to radiation was likely due to genetic differences in the capacity to respond to environmental challenges between these two species (Liu et al., 2013). Thus, understanding the interaction between genetic risk factors and bio-protective measures can help us to limit, and potentially counter, the negative impacts that space travel can have on astronauts and future space tourists (Schmidt and Goodwin, 2013).

Most effects of space flight go back to normal a while after return to Earth, for example, the changes of mtDNA gene expression that occurred during spaceflight in the Twin Study returned to baseline levels already within a few weeks post-flight. However, many astronauts after returning from the ISS present health issues similar to geriatric stress, including immune, endocrine, central nervous system, and cardiovascular changes, muscle sarcopenia, osteoporosis, and anemia, which effects are associated with dysfunction in mitochondria, suggesting that impact of space flight resemble accelerated ageing (Gao et al., 2023; Rubinstein et al., 2021; Yatagai et al., 2019). As most of those ageing-like changes associated with spaceflight are reversible, whether some effects can be long-term or even life-long remains unclear. Another yet poorly understood health concern of long and multigenerational space travel is the effects of space hazards on reproduction and fetal development. A developing fetus with its elevated growth rate, is especially at risk of increased DNA damage and genetic mutations resulting from space radiation. Moreover, oxidative stress has been linked to placental dysfunctions, which can lead to pre-term birth and several metabolic disorders (Jain et al., 2023). Therefore, expected prolonged missions to the Moon and Mars pose yet unknown health challenges that may result in permanent pathological impairments.

10. Strategies for protecting mitochondrial function in spaceflight

Oxidative stress induced by ionizing radiation and microgravity in space is linked to mitochondrial dysregulation, DNA mutations, altered cellular functions and carcinogenesis. Several countermeasure strategies have been discussed with the potential to help combat the alterations to normal mitochondrial functions in spaceflight. Such promising strategies include developing natural methods of protective shielding from ionizing radiation, the study of extremotolerant organisms to identify their protective mechanisms, identifying epigenetic changes and individual genetic variability as means to assess individual health risks and personalised medicine strategies, dietary supplementation with exogenous antioxidants, and combating circadian dysregulation (Seoane-Viano et al., 2022; Pavez Lorie et al., 2021; Gómez et al., 2021; Malhan et al., 2023).

Some innovative approaches to combating the oxidative stress caused by space radiation involve the use of natural or synthetic compounds that absorb and protect from radiation, such as melanin and selenium (Seoane-Viano et al., 2022). The combination of melanin with selenium led to the synthesis of selenomelanin, a compound that absorbs radiation more efficiently than melanin alone. Thus, the discovery of more compounds with similar properties could help to protect astronauts during long-term space missions, however more research is needed. Another exciting approach involves the identification of unique compounds that combat oxidative stress in extremophile organisms. For example, extremophile tardigrades possess a unique damage suppressor protein that can protect the animal from free radicals produced by ionizing radiation. The protective effect of this protein could potentially be used to mitigate oxidative damage resulting from space radiation in humans. Identifying similar bioactive compounds is a current focus of extensive research (Seoane-Viano et al., 2022).

Mitochondrial dysregulation and oxidative stress in spaceflight is linked to epigenetic changes, including altered methylation patterns, but also gene and protein expression changes, and genetic mutations. This highlights the importance of monitoring each crew member to assess their health risks, as a means of developing personalized countermeasures (Seoane-Viano et al., 2022). Individualized medicine depends on reliable diagnostic tools which can be used for the selection of treatment solutions. Genetic and epigenetic factors determine what makes the individual more vulnerable to the effects of spaceflight, especially space radiation. Therefore, the concept of personalised precision medicine for astronauts implies that specific gene mutations, gene expression patterns, and biomarkers should be studied to identify radiation-sensitive and radiation-resistant individuals before space missions. Novel biomarkers will soon allow for a routine identification of individuals with higher radiation sensitivity. Biomarkers based on new microfluidics and lab-on-chip technology can also measure ROS and endogenous antioxidant production during spaceflight (Gómez et al., 2021). Thus, personalized space medicine is becoming increasingly important in context of long space missions to the Moon and Mars (Pavez Lorie et al., 2021).

Another promising approach directly associated with the concept of personalized medicine is supplementation with antioxidant dietary compounds. Antioxidants play a protective role by reducing oxidative

damage to lipids, nucleic acids, and proteins. However, strategies to counteract the detrimental effects of oxidative stress in spaceflight by supplementing anti-oxidative cocktails have yet been unsuccessful. This could be caused by a lack of prior genetic testing that determines the capacity to produce endogenous antioxidants by each astronaut, emphasizing the importance of an individualized approach to dietary supplementation (Pavez Lorie et al., 2021; Gómez et al., 2021). Moreover, exposure to the space environment leads to physiological changes that can affect the pharmacokinetics of some medicines and dietary compounds (Pavez Lorie et al., 2021). Thus, identifying genetic variants that play a role in the predisposition to higher oxidative damage is key to the effective administration of antioxidant-cocktail preventive strategies (Gómez et al., 2021).

Antioxidants can be classified into two broad categories: endogenous and exogenous. Our body produces natural antioxidants such as superoxide dismutase, catalase, and glutathione peroxidase, which help to maintain a healthy redox balance. But, when ROS levels increase excessively, exogenous antioxidants may be beneficial. However, while ROS are detrimental in excess, leading to oxidative damage to lipids, proteins, DNA, and mitochondrial stress, they have a physiological role in cell differentiation, immune activation, and autophagy. Therefore, while an ideal antioxidant mix should reduce ROS, it must ensure that an optimal amount of ROS remains to perform cellular signalling functions. However the antioxidant protective properties against ROS vary according to the genetics of the individual, and when administered on Earth versus spaceflight. Thus, the different properties of each antioxidant and relative ROS exposure should be taken into account to create a supplement cocktail that is optimal for each individual (Gómez et al., 2021). The most commonly used exogenous dietary antioxidants are vitamin C, E, A, luteolin, resveratrol, and coenzyme Q10. Resveratrol is a polyphenol found in grapes, blueberries, and wine, with potent antioxidant and anti-inflammatory properties against oxidative stress and mitochondrial dysfunction. Resveratrol attenuates the cytotoxicity of H_2O_2 by its oxygen-scavenging hydroxyl functional groups. Coenzyme Q10 (ubiquinone) reduces ROS production in mitochondria. It plays an important role in mitochondrial ETS, as it accepts and transfers electrons between complexes I and III. It prevents damage to DNA and proteins, lipid peroxidation, stabilizes calcium channels, activates uncoupling proteins, and improves mitochondrial biogenesis (Gómez et al., 2021).

An additional approach to protect mitochondrial functions in spaceflight is to combat circadian dysregulation. In mammals, about half of the transcriptome is rhythmically expressed with daily or shorter oscillations. The circadian clock regulates the expression and activity of mitochondrial metabolic and antioxidant enzymes. However, circadian dysregulation is also a hallmark of terrestrial ageing and space-related ageing in astronauts (Malhan et al., 2023). Aging is associated with dysregulation of several cellular processes including mitochondrial dysfunction, genomic instability, and epigenetic alterations. Moreover, space conditions alter circadian clock genes, and several of these genes are major regulatory genes affecting upregulation of pro-inflammatory pathways and downregulation of anti-oxidative pathways. Inflammation and oxidative stress cause physiological and mitochondrial function impairments in spaceflight. Moreover, a recent comprehensive analysis showed common gene expression changes in the circadian clock and skeletal muscle due to ageing, both on Earth and in long-term spaceflight (Malhan et al., 2023). Thus, using drugs such as melatonin could be beneficial in re-establishing the proper circadian system functions in extended spaceflight missions (Seoane-Viano et al., 2022).

11. Conclusions

Mitochondria are cellular organelles crucial to many physiological processes that are known to be affected by spaceflight. This paper briefly summarizes how mitochondrial function is affected by all the main health hazards associated with spaceflight, including the most obvious being ionizing radiation and microgravity, but also the more subtle ones being non-ionizing radiation, confinement, or circadian cycles (Fig. 1). Mitochondria in space are affected at several critical levels including morphological, functional, genomic, and proteomic, leading to a variety of physiological effects on animal energetics, and additional effects associated with mitochondrial oxidative stress. Spaceflight leaves the organism with a range of oxidative stress-related changes, some of which resemble senescence. Therefore, a growing number of researchers regard spaceflight as a model of accelerated ageing and a model of mitochondrial disorders. This perspective calls for consideration whether possibly common treatments to mitigate the effects of senescence and spaceflight could be developed to help achieve both healthy ageing and safe spaceflight.

As the current long-term space missions last only about a year, the mitochondria-related impairments usually are not permanent, and the astronauts' health goes back to normal a while after returning to Earth. However, the long-term exploration of the Moon and Mars will endanger the astronaut's health for much longer, without the chance of quick recovery in Earthly conditions. The future plans of colonizing the Moon and Mars may mean that the astronauts will live in space conditions for many generations. At the moment, the impact of such long-term or even multi-generational ventures is unknown. Uncertain is also how to address the growing interest in space tourism, which means that the average passenger, that may not be of exceptional health or physical fitness, unlike the selected highly-trained astronauts, would also have to be provided with a safe spaceflight experience. We argue that investigating mitochondrial biology in space conditions can help to resolve many of these challenges of spaceflight and that mitochondria should be the focus of space health studies.

CRediT authorship contribution statement

Agata M. Rudolf: Conceptualization, Writing – original draft, Writing – review & editing, Visualization, Investigation. **Wendy R. Hood:** Funding acquisition, Writing – review & editing, Supervision.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Acknowledgements

We would like to thank the GeneLab Animals and Multiomics AWGs members, and the TRISH B-SURE program for their inspiration and support during several group seminars, workshops and interesting discussions.

Funding

This work has been supported by a National Science Foundation award to WR Hood, OIA1736150.

References

Afshinnekoo, E., Scott, R.T., MacKay, M.J., Pariset, E., Cekanaviciute, E., Barker, R., Gilroy, S., Hassane, D., Smith, S.M., Zwart, S.R., Nelman-Gonzalez, M., 2020. Fundamental biological features of spaceflight: advancing the field to enable deep-space exploration. Cell 183 (5), 1162–1184.

Barjaktarovic, Z., Schmaltz, D., Shyla, A., Azimzadeh, O., Schulz, S., Haagen, J., Dörr, W., Sarioglu, H., Schäfer, A., Atkinson, M.J., Zischka, H., 2011. Radiation–induced signaling results in mitochondrial impairment in mouse heart at 4 weeks after exposure to X-rays. PLoS One 6 (12), 27811.

Barneche, D.R., Jahn, M., Seebacher, F., 2019. Warming increases the cost of growth in a model vertebrate. Funct. Ecol. 33 (7), 1256–1266.

Bratic, I., Trifunovic, A., 2010. Mitochondrial energy metabolism and ageing. Biochim. Biophys. Acta Bioenerg. 1797 (6), 961–967.

Chakraborty, N., Waning, D.L., Gautam, A., Hoke, A., Sowe, B., Youssef, D., Butler, S., Savaglio, M., Childress, P.J., Kumar, R., Moyler, C., 2020. Gene-metabolite network

linked to inhibited bioenergetics in association with spaceflight-induced loss of male mouse quadriceps muscle. J. Bone Miner. Res. 35 (10), 2049–2057.
Coleman, M.A., Sasi, S.P., Onufrak, J., Natarajan, M., Manickam, K., Schwab, J., Muralidharan, S., Peterson, L.E., Alekseyev, Y.O., Yan, X., Goukassian, D.A., 2015. Low-dose radiation affects cardiac physiology: gene networks and molecular signaling in cardiomyocytes. Am. J. Physiol. Heart Circul. Physiol. 309 (11), 1947–1963.
Connor, M.K., Hood, D.A., 1998. Effect of microgravity on the expression of mitochondrial enzymes in rat cardiac and skeletal muscles. J. Appl. Physiol. 84 (2), 593–598.
Curley, S.A., Palalon, F., Lu, X., Koshkina, N.V., 2014. Noninvasive radiofrequency treatment effect on mitochondria in pancreatic cancer cells. Cancer 120 (21), 3418–3425.
Destefanis, M., Viano, M., Leo, C., Gervino, G., Ponzetto, A., Silvagno, F., 2015. Extremely low frequency electromagnetic fields affect proliferation and mitochondrial activity of human cancer cell lines. Int. J. Radiat Biol. 91 (12), 964–972.
Esmekaya, M., Canseven Kursun, A., Kayhan, H., Tuysuz, M., Sirav, B., Seyhan, N., 2017. Mitochondrial hyperpolarization and cytochrome-c release in microwave-exposed MCF-7 cells. Gen. Physiol. Biophys. 36 (2).
Finkel, T., Holbrook, N.J., 2000. Oxidants, oxidative stress and the biology of ageing. Nature 408 (6809), 239–247.
Gao, T., Huang, J., Zhang, X., Gao, F., 2023. Exercise counteracts vascular aging in long-term spaceflight: challenges and perspective. Curr. Opin. Physiol. 100628.
Garrett-Bakelman, F.E., Darshi, M., Green, S.J., Gur, R.C., Lin, L., Macias, B.R., McKenna, M.J., Meydan, C., Mishra, T., Nasrini, J., Piening, B.D., 2019. The NASA Twins Study: A multidimensional analysis of a year-long human spaceflight. Science 364 (6436), 8650.
Gómez, X., Sanon, S., Zambrano, K., Asquel, S., Bassantes, M., Morales, J.E., Otáñez, G., Pomaquero, C., Villarroel, S., Zurita, A., Calvache, C., 2021. Key points for the development of antioxidant cocktails to prevent cellular stress and damage caused by reactive oxygen species (ROS) during manned space missions. Microgravity 7 (1), 35.
Hao, Y.H., Zhang, J., Wang, H., Wang, H.Y., Dong, J., Xu, X.P., Yao, B.W., Wang, L.F., Zhou, H.M., Zhao, L., Peng, R.Y., 2018. HIF-1α regulates COXIV subunits, a potential mechanism of self-protective response to microwave induced mitochondrial damages in neurons. Sci. Rep. 8 (1), 10403.
Heine, K.B., Justyn, N.M., Hill, G.E., Hood, W.R., 2021. Ultraviolet irradiation alters the density of inner mitochondrial membrane and proportion of inter-mitochondrial junctions in copepod myocytes. Mitochondrion 56, 82–90.
Hollis, F., Pope, B.S., Gorman-Sandler, E., Wood, S.K., 2022. Neuroinflammation and mitochondrial dysfunction link social stress to depression. Neurosci. Soc. Stress 59–93.
Hood, W.R., Zhang, Y., Mowry, A.V., Hyatt, H.W., Kavazis, A.N., 2018. Life history trade-offs within the context of mitochondrial hormesis. Integr. Comp. Biol. 58 (3), 567–577.
Indo, H.P., Majima, H.J., Terada, M., Suenaga, S., Tomita, K., Yamada, S., Higashibata, A., Ishioka, N., Kanekura, T., Nonaka, I., Hawkins, C.L., 2016. Changes in mitochondrial homeostasis and redox status in astronauts following long stays in space. Sci. Rep. 6 (1), 39015.
Jain, V., de Sousa, C., Lopes, S.M., Benotmane, M.A., Verratti, V., Mitchell, R.T., Stukenborg, J.B., 2023. Human development and reproduction in space-A European perspective. Microgravity 9 (1), 24.
Jeong, A.J., Kim, Y.J., Lim, M.H., Lee, H., Noh, K., Kim, B.H., Chung, J.W., Cho, C.H., Kim, S., Ye, S.K., 2018. Microgravity induces autophagy via mitochondrial dysfunction in human Hodgkin's lymphoma cells. Sci. Rep. 8 (1), 14646.
Ji, G., Chang, H., Yang, M., Chen, H., Wang, T., Liu, X., Lv, K., Li, Y., Song, B., Qu, L., 2022. The mitochondrial proteomic changes of rat hippocampus induced by 28-day simulated microgravity. PLoS One 17 (3), 0265108.
Kim, D.S., Weber, T., Straube, U., Hellweg, C.E., Nasser, M., Green, D.A., Fogtman, A., 2021. The potential of physical exercise to mitigate radiation damage- A systematic review. Front. Med. 8, 393.
Koch, R.E., Buchanan, K.L., Casagrande, S., Crino, O., Dowling, D.K., Hill, G.E., Hood, W. R., McKenzie, M., Mariette, M.M., Noble, D.W., Pavlova, A., 2021. Integrating mitochondrial aerobic metabolism into ecology and evolution. Trends Ecol. Evol. 36 (4), 321–332.
Liu, W., Haley, B., Kwasny, M.J., Li, J.J., Grdina, D.J., Paunesku, T. and Woloschak, G.E. (2013). Comparing radiation toxicities across species: an examination of radiation effects in Mus musculus and Peromyscus leucopus. *International Journal of Radiation Biology*, 2013.
Liu, P., Li, D., Li, W., Wang, D., 2019. Mitochondrial unfolded protein response to microgravity stress in nematode Caenorhabditis elegans. Sci. Rep. 9 (1), 1–9.
Livingston, K., Schlaak, R.A., Puckett, L.L., Bergom, C., 2020. The role of mitochondrial dysfunction in radiation-induced heart disease: from bench to bedside. Front. Cardiovasc. Med. 7, 20.
Locatelli, L., Cazzaniga, A., De Palma, C., Castiglioni, S., Maier, J., 2020. Mitophagy contributes to endothelial adaptation to simulated microgravity. FASEB J. 34 (1), 1833–1845.
Luxton, J.J., McKenna, M.J., Taylor, L.E., George, K.A., Zwart, S.R., Crucian, B.E., Drel, V.R., Garrett-Bakelman, F.E., Mackay, M.J., Butler, D., Foox, J., 2020. Temporal telomere and DNA damage responses in the space radiation environment. Cell Rep. 33 (10), 108435.
Malhan, D., Schoenrock, B., Yalçin, M., Blottner, D., Relógio, A., 2023. Circadian regulation in aging: Implications for spaceflight and life on earth. Aging Cell 22 (9), 13935.
Mao, X.W., Crapo, J.D., Gridley, D.S., 2012. Mitochondrial oxidative stress-induced apoptosis and radioprotection in proton-irradiated rat retina. Radiat. Res. 178 (3), 118–125.
Mao, X.W., Nishiyama, N.C., Pecaut, M.J., Campbell-Beachler, M., Gifford, P., Haynes, K. E., Becronis, C., Gridley, D.S., 2016. Simulated microgravity and low-dose/low-dose-rate radiation induces oxidative damage in the mouse brain. Radiat. Res. 185 (6), 647–657.
Mao, X.W., Nishiyama, N.C., Campbell-Beachler, M., Gifford, P., Haynes, K.E., Gridley, D. S., Pecaut, M.J., 2017. Role of NADPH oxidase as a mediator of oxidative damage in low-dose irradiated and hindlimb-unloaded mice. Radiat. Res. 188 (4), 392–399.
Maremonti, E., Eide, D.M., Rossbach, L.M., Lind, O.C., Salbu, B., Brede, D.A., 2020. In vivo assessment of reactive oxygen species production and oxidative stress effects induced by chronic exposure to gamma radiation in Caenorhabditis elegans. Free Radical Biol. Med. 152, 583–596.
McDonald, J.T., Stainforth, R., Miller, J., Cahill, T., Silveira, W.A.D., Rathi, K.S., Hardiman, G., Taylor, D., Costes, S.V., Chauhan, V., Meller, R., 2020. NASA genelab platform utilized for biological response to space radiation in animal models. Cancers 12 (2), 381.
Michaletti, A., Gioia, M., Tarantino, U., Zolla, L., 2017. Effects of microgravity on osteoblast mitochondria: a proteomic and metabolomics profile. Sci. Rep. 7 (1), 1–12.
Monzel, A.S., Enríquez, J.A., Picard, M., 2023. Multifaceted mitochondria: moving mitochondrial science beyond function and dysfunction. Nature Metabolism 5 (4), 546–562.
Naviaux, R.K., 2019. Metabolic features and regulation of the healing cycle-A new model for chronic disease pathogenesis and treatment. Mitochondrion 46, 278–297.
Nelson, E.S., Mulugeta, L., Myers, J.G., 2014. Microgravity-induced fluid shift and ophthalmic changes. Life 4 (4), 621–665.
Nguyen, H.P., Tran, P.H., Kim, K.S., Yang, S.G., 2021. The effects of real and simulated microgravity on cellular mitochondrial function. npj Microgravity 7 (1), 44.
Nugent, S., Mothersill, C.E., Seymour, C., McClean, B., Lyng, F.M., Murphy, J.E., 2010. Altered mitochondrial function and genome frequency post exposure to γ-radiation and bystander factors. Int. J. Radiat Biol. 86 (10), 829–841.
Pavez Lorie, E., Baatout, S., Choukér, A., Buchheim, J.I., Baselet, B., Dello Russo, C., Wotring, V., Monici, M., Morbidelli, L., Gagliardi, D., Stingl, J.C., 2021. The future of personalized medicine in space: From observations to countermeasures. Front. Bioeng. Biotechnol. 9, 739747.
Picard, M., McEwen, B.S., 2018. Psychological stress and mitochondria: a systematic review. Psychosom. Med. 80 (2), 141.
Powers, S.K., Radak, Z., Ji, L.L., 2016. Exercise-induced oxidative stress: past, present and future. J. Physiol. 594 (18), 5081–5092.
Raber, J., Holden, S., Sudhakar, R., Hall, R., Glaeser, B., Lenarczyk, M., Rockwell, K., Nawarawong, N., Sterrett, J., Perez, R., Leonard, S.W., 2021. Effects of 5-ion beam irradiation and hindlimb unloading on metabolic pathways in plasma and brain of behaviorally tested WAG/RIJ rats. Front. Physiol. 12.
Radstake, W.E., Gautam, K., Miranda, S., Vermeesen, R., Tabury, K., Rehnberg, E., Buset, J., Janssen, A., Leysen, L., Neefs, M., Verslegers, M., 2023. The effects of combined exposure to simulated microgravity, ionizing radiation, and cortisol on the in vitro wound healing process. Cells 12 (2), 246.
Reitz, G., Berger, T., Matthiae, D., 2012. Radiation exposure in the moon environment. Planet. Space Sci. 74 (1), 78–83.
Richardson, R.B., Mailloux, R.J., 2023. Mitochondria need their sleep: redox, bioenergetics, and temperature regulation of circadian rhythms and the role of cysteine-mediated redox signaling, uncoupling proteins, and substrate cycles. Antioxidants 12 (3), 674.
Rubinstein, L., Schreurs, A.S., Torres, S.M., Steczina, S., Lowe, M.G., Kiffer, F., Allen, A. R., Ronca, A.E., Sowa, M.B., Globus, R.K., Tahimic, C.G., 2021. Overexpression of catalase in mitochondria mitigates changes in hippocampal cytokine expression following simulated microgravity and isolation. npj Microgravity 7 (1), 24.
Salin, K., Auer, S.K., Rey, B., Selman, C., Metcalfe, N.B., 2015. Variation in the link between oxygen consumption and ATP production, and its relevance for animal performance. Proc. R. Soc. B Biol. Sci. 282 (1812), 20151028.
Salin, K., Auer, S.K., Rudolf, A.M., Anderson, G.J., Selman, C., Metcalfe, N.B., 2016. Variation in metabolic rate among individuals is related to tissue-specific differences in mitochondrial leak respiration. Physiol. Biochem. Zool. 89 (6), 511–523.
Schatten, H., Lewis, M.L., Chakrabarti, A., 2001. Spaceflight and clinorotation cause cytoskeleton and mitochondria changes and increases in apoptosis in cultured cells. Acta Astronaut. 49 (3–10), 399–418.
Schmidt, M.A., Goodwin, T.J., 2013. Personalized medicine in human space flight: using Omics based analyses to develop individualized countermeasures that enhance astronaut safety and performance. Metabolomics 9 (6), 1134–1156.
Seoane-Viano, I., Ong, J.J., Basit, A.W., Goyanes, A., 2022. To infinity and beyond: strategies for fabricating medicines in outer space. Int. J. Pharm.: X 4, 100121.
Silveira, W.A., Fazelinia, H., Rosenthal, S.B., Laiakis, E.C., Kim, M.S., Meydan, C., Kidane, Y., Rathi, K.S., Smith, S.M., Stear, B., Ying, Y., 2020. Comprehensive multi-omics analysis reveals mitochondrial stress as a central biological hub for spaceflight impact. Cell 183 (5), 1185–1201.
Simmons, P., Trujillo, M., McElroy, T., Binz, R., Pathak, R., Allen, A.R., 2022. Evaluating the effects of low-dose simulated galactic cosmic rays on murine hippocampal-dependent cognitive performance. Front. Neurosci. 16, 1688.
Simonsen, L.C., Slaba, T.C., Guida, P., Rusek, A., 2020. NASA'S first ground-based Galactic Cosmic Ray Simulator: enabling a new era in space radiobiology research. PLoS Biol. 18 (5), 3000669.
Singh, R., Rajput, M., Singh, R.P., 2021. Simulated microgravity triggers DNA damage and mitochondria-mediated apoptosis through ROS generation in human promyelocytic leukemic cells. Mitochondrion 61, 114–124.

Sokolova, I., 2018. Mitochondrial adaptations to variable environments and their role in animals' stress tolerance. Integr. Comp. Biol. 58 (3), 519–531.
Speakman, J.R., Talbot, D.A., Selman, C., Snart, S., McLaren, J.S., Redman, P., Krol, E., Jackson, D.M., Johnson, M.S., Brand, M.D., 2004. Uncoupled and surviving: individual mice with high metabolism have greater mitochondrial uncoupling and live longer. Aging Cell 3 (3), 87–95.
Tahimic, C.G., Paul, A.M., Schreurs, A.S., Torres, S.M., Rubinstein, L., Steczina, S., Lowe, M., Bhattacharya, S., Alwood, J.S., Ronca, A.E., Globus, R.K., 2019. Influence of social isolation during prolonged simulated weightlessness by hindlimb unloading. Front. Physiol. 10, 1147.
Takeda, M., Magaki, T., Okazaki, T., Kawahara, Y., Manabe, T., Yuge, L., Kurisu, K., 2009. Effects of simulated microgravity on proliferation and chemosensitivity in malignant glioma cells. Neurosci. Lett. 463 (1), 54–59.
Thomas, L.W., Ashcroft, M., 2019. Exploring the molecular interface between hypoxia-inducible factor signalling and mitochondria. Cell. Mol. Life Sci. 76, 1759–1777.
Treberg, J.R., Munro, D., Jastroch, M., Quijada-Rodriguez, A.R., Kutschke, M., Wiens, L., 2018. Comparing electron leak in vertebrate muscle mitochondria. Integr. Comp. Biol. 58 (3), 495–505.
Vernice, N.A., Meydan, C., Afshinnekoo, E., Mason, C.E., 2020. Long-term spaceflight and the cardiovascular system. Precis. Clin. Med. 3 (4), 284–291.
Walton, J.C., Weil, Z.M., Nelson, R.J., 2011. Influence of photoperiod on hormones, behavior, and immune function. Front. Neuroendocrinol. 32 (3), 303–319.
Wang, Y., Javed, I., Liu, Y., Lu, S., Peng, G., Zhang, Y., Qing, H., Deng, Y., 2016. Effect of prolonged simulated microgravity on metabolic proteins in rat hippocampus: steps toward safe space travel. J. Proteome Res. 15 (1), 29–37.
Yatagai, F., Honma, M., Dohmae, N., Ishioka, N., 2019. Biological effects of space environmental factors: a possible interaction between space radiation and microgravity. Life Sci. Space Res. 20, 113–123.
Yin, Z., Yang, G., Deng, S., Wang, Q., 2019. Oxidative stress levels and dynamic changes in mitochondrial gene expression in a radiation-induced lung injury model. J. Radiat. Res. 60 (2), 204–214.
Zhang, Y., Humes, F., Almond, G., Kavazis, A.N., Hood, W.R., 2018. A mitohormetic response to pro-oxidant exposure in the house mouse. Am. J. Physiol.-Regul. Integr. Comp. Physiol. 314 (1), 122–134.
Zhang, R., Ran, H.H., Cai, L.L., Zhu, L., Sun, J.F., Peng, L., Liu, X.J., Zhang, L.N., Fang, Z., Fan, Y.Y., Cui, G., 2014. Simulated microgravity-induced mitochondrial dysfunction in rat cerebral arteries. FASEB J. 28 (6), 2715–2724.
Zheng, H.X., Tian, W.M., Yan, H.J., Jiang, H.D., Liu, S.S., Yue, L., Han, F., Wei, L.J., Chen, X.B., Li, Y., 2012. Expression of estrogen receptor α in human breast cancer cells regulates mitochondrial oxidative stress under simulated microgravity. Adv. Space Res. 49 (10), 1432–1440.
Zuo, L., Zhou, T., Pannell, B.K., Ziegler, A.C., Best, T.M., 2015. Biological and physiological role of reactive oxygen species–the good, the bad and the ugly. Acta Physiol. 214 (3), 329–348.