Fiscal Year:	FY 2021	**Task Last Updated:**	FY 08/25/2021
PI Name:	Lawler, John Ph.D.		
Project Title:	Attenuation of Space Radiation-induced Pro-oxidant and Fibrotic Signaling in the Heart by Nutritional and Genetic Interventions: Adventures in Tissue Sharing		
Division Name:	Human Research		
Program/Discipline:			
Program/Discipline--Element/Subdiscipline:			
Joint Agency Name:		**TechPort:**	No
Human Research Program Elements:	(1) **SR**:Space Radiation		
Human Research Program Risks:	(1) **Cardiovascular**:Risk of Cardiovascular Adaptations Contributing to Adverse Mission Performance and Health Outcomes		
Space Biology Element:	None		
Space Biology Cross-Element Discipline:	None		
Space Biology Special Category:	None		
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PI Organization Type:	UNIVERSITY	**Phone:**	979-862-2038
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Zip Code:	77843	**Congressional District:**	17
Comments:			
Project Type:	Ground	**Solicitation / Funding Source:**	2016-2017 HERO NNJ16ZSA001N-Crew Health (FLAGSHIP, OMNIBUS). Appendix A-Omnibus, Appendix B-Flagship
Start Date:	07/01/2017	**End Date:**	06/30/2022
No. of Post Docs:	1	**No. of PhD Degrees:**	
No. of PhD Candidates:	2	**No. of Master' Degrees:**	
No. of Master's Candidates:	1	**No. of Bachelor's Degrees:**	2
No. of Bachelor's Candidates:	7	**Monitoring Center:**	NASA JSC
Contact Monitor:	Zawaski, Janice	**Contact Phone:**	
Contact Email:	janice.zawaski@nasa.gov		
Flight Program:			
Flight Assignment:	NOTE: End date changed to 6/30/2022 per Space Radiation element and NSSC information (Ed., 7/6/21) NOTE: End date changed to 6/30/2021 per NSSC information (Ed., 11/6/20) NOTE: End date changed to 6/30/2020 per NSSC information (Ed., 9/26/19)		
Key Personnel Changes/Previous PI:	July 2021 report: Adding Mariam Atef, PhD student. Adding: Amelia Flug (Aggie Research Scholar) 2021- ; Devon Roeming (Aggie Research Scholar) 2021- ; Corine Harvey (Aggie Research Scholar) 2021- ; Grace Barrow (Aggie Research Scholar) 2021- ; Samhitha Ramanuja (Aggie Research Scholar) 2021- ; Binh Nguyen (Aggie Research Scholar) 2021- . REMOVED due to COVID: Jordyn Johnson, MS student; Myles McFarland (Aggie Research Scholar) 2020; Mollie Linder (Aggie Research Scholar) 2020; Hallie Harris (Aggie Research Scholar) 2020; Aakash Kothari (Aggie Research Scholar) 2020; Francisco Melesio (Aggie Research Scholar) 2020; Sonny Rodriguez (Aggie Research Scholar) 2020; Mia Ngyuen (Aggie Research Scholar) 2020.		
COI Name (Institution):	Ford, John Ph.D. (Texas A&M Engineering Experiment Station) Turner, Nancy Ph.D. (Texas A&M AgriLife Research)		
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Performance Goal No.:	
Performance Goal Text:	
Task Description:	Spaceflight imposes a unique set of stressors on astronauts as a result of mechanical unloading due to microgravity conditions, while tissues are bombarded by galactic and solar radiation. The cardiovascular system is adversely affected by the disuse and fluid shifts that occur with spaceflight. However, there is a growing concern that cardiovascular disease may be substantially elevated during spaceflight. Indeed, increasing evidence indicates that radiation exposure causes damage and fibrosis in the heart and vasculature. Cellular mechanisms of dysfunction due to disuse and space radiation include increased oxidative stress, pro-inflammatory signaling, and impaired function. Heart, vasculature, and the musculoskeletal system will be exposed to gamma and heavy ion (HZE) radiation. Mitochondria, lysosomes, and nucleic acids are particularly susceptible to HZE and secondary oxidant-induced damage. Previous findings and preliminary data from our laboratory indicate that oxidative stress contributes to apoptosis and fibrosis in aging heart models. However, the contribution by which space radiation (X-Ray, HZE) contributes to secondary oxidative stress and fibrosis in the heart are not well understood. We argue that space radiation induced acceleration of the aging process in heart and skeletal muscle, where susceptibility to fibrosis and apoptosis is high. New studies and Preliminary Data from our laboratory suggest that the renin-angiotensin signaling (RAS) are significant sources of oxidative stress, and thus pro-fibrotic signaling in the heart. Upregulation of RAS in the aging heart upregulates the Nox2 isoform of NADPH oxidase. We have also recently found that Nox2 contributes to oxidative stress and atrophy during ground-based spaceflight of skeletal muscle. Thus secondary and amplified oxidative stress may damage nuclei and stimulate pro-fibrotic signaling, including TGF-ß, smad2/3 phosphorylation, and collagen I accumulation. The current RFA research emphasis in Space Biology Tissue Sharing provides an opportunity to promote sharing of samples with ongoing and archived studies. We will propose a series of studies with X-Ray, HZE, and X-Ray + HZE radiation. Collaboration with Dr. Nancy Turner's laboratory at Texas A&M University will focus on two sets of radiation studies. The first cohort of studies will use X-Ray radiation (0.5 Gy) to induce damage and oxidative stress. Mouse (astronaut age) heart samples will be taken 12 hours, or 4 or 8 weeks after exposure. In the second set of experiments, mice will be exposed to 28Si and 48Ti (0.5 Gy). Mice will be sacrificed and tissues extracted 12 hrs, 4 wks, or 8 wks after radiation exposure. Efficacy of an intervention of fish oil + pectin in reducing cardiac fibrotic signaling will be tested. Fish oil reduces oxidative stress and cardiovascular disease, increases protective heat shock proteins. Our Preliminary Data reveal that fish oil + curcumin also reduces muscle atrophy. Dietary pectin ingestion reduces oxidative stress and apoptosis. Pectin and fish oil have also reduced radiation-induced tissue fibrosis in the kidney and liver, respectively. However, the effects on the irradiated heart are unknown. p53 contributes to apoptosis, cardiac fibrosis, and muscle atrophy. We will also query archived cardiac samples irradiated at the Brookhaven National Laboratory involved in combined X-Ray and 56Fe radiation, where mice with a single p53 allele deletion and wild-types were irradiated.
Rationale for HRP Directed Research:	
Research Impact/Earth Benefits:	The cardiovascular system experiences a number of dynamic changes during spaceflight that impair function and predispose it to chronic disease. When space missions travel beyond the protection of the Van Allen belts the hearts and vasculature of astronauts are subject to the profound stressors of both microgravity and radiation from solar and galactic sources. Mechanical unloading of the musculoskeletal system due to microgravity results in severe disuse, eliciting "detraining" of the heart. In addition, a fluid shift toward central blood volume during microgravity results in elevated right atrial pressure and thus elimination of plasma volume via diuresis. Atrial naturietic factor (ANF) and the renin-angiotensin II pathway are involved in increased renal excretion of water. Spaceflight appears to elicit morphological (e.g., collagen fibrosis) and functional changes of the heart that could impede performance, lead to fatigue and orthostatic hypotension upon re-entry to a gravitational environment, and increase the risk of heart and vascular disease. In addition, disuse that occurs with microgravity may predispose the heart to arrhythmias (Moffitt et al. 2013). Radiation enhances apoptosis and loss of myocytes as well as accumulation of collagenous tissue, or "fibrosis." The average age of a typical astronaut has increased to over 50 years of age, and progressive age increases oxidative stress in the heart (Kwak et al. 2006). Spaceflight imposes a unique set of stressors on astronauts as a result of the loss of gravity during spaceflight, while tissues are bombarded by galactic and solar radiation. The cardiovascular system is adversely affected by the disuse and fluid shifts that occur with spaceflight. However, there is a growing concern that cardiovascular disease may be substantially elevated during spaceflight. Indeed, increasing evidence indicates that radiation exposure causes damage and fibrosis in the heart and vasculature. Weightlessness and space radiation during long-duration spaceflight, particularly in outer space between the Earth and the moon or Mars, increases inflammation and oxidative stress in the heart, vasculature, and muscles, joints, and bones. The body is exposed to X-ray and heavy ion (HZE) radiation that damages cell components such as mitochondria, nuclei, and the cell membrane through increase release of oxidants (i.e., oxidative stress). Astronaut age has increased into the 50s, and thus has the risk of damage, cell death, and fibrotic connective tissue, as published by our laboratory and other scientists. However, the contribution by which space radiation (X-Ray, HZE) contributes to secondary oxidative stress and fibrosis in the heart is poorly understood. We argue that space radiation accelerated the aging process in heart and skeletal muscle, increased fibrosis, and contributed to cell death. New publications and pilot data from our laboratories indicate that a potential source of oxidative stress in the heart during radiation is called the renin-angiotensin system (RAS). RAS can trigger the assembly of NADPH oxidase-2 (Nox2), a cluster of proteins that produces oxidative stress. We recently found that Nox-2 is elevated in a ground spaceflight analog in skeletal muscle and heart, and contributed directly to changes in muscle cell size, shape, and infiltration of connective tissue. Antioxidant compounds and nutritional supplement choices that are based upon causal studies may have alleviated changes in the heart, vasculature, and skeletal muscle with spaceflight. For example, fish oil reduces oxidative stress, and thus increases protective heat shock proteins, and reduces cardiovascular disease. For example, a combination of fish oil and curcumin recently prevented muscle fiber atrophy and increased protective stress response proteins in a spaceflight analog. Dietary pectin ingestion reduces oxidative stress and cell death. Pectin and fish oil have also reduced radiation-induced tissue fibrosis in the kidney and liver, respectively. However, the effects on the irradiated heart are unknown. We propose to determine the effects of a combination of fish oil and pectin on heavy ion-induced radiation in the heart.

	The current RFA research emphasis in Space Biology Tissue Sharing provides an opportunity to promote sharing of samples with ongoing and archived studies. We are conducting a series of studies with X-Ray, HZE, and X-Ray + HZE radiation. Collaboration with Dr. Nancy Turner's laboratory at Texas A&M University focuses on two sets of radiation studies. The first cohort of studies will use X-Ray radiation (0.5 Gy) to induce damage and oxidative stress. Mouse (astronaut age) heart samples will be taken 12 hours, or 4 or 8 weeks after exposure. In the second set of experiments, mice will be exposed to 28Si and 48Ti (0.5 Gy). Mice were sacrificed and tissues extracted 12 hrs, 4 wks, or 8 wks after radiation exposure. Effectiveness of fish oil + pectin in reducing heart damage and fibrosis is being tested. Our Preliminary Data reveal that fish oil + curcumin also reduces muscle atrophy. A protein called p53 also contributes to cell death, fibrosis of the heart, and muscle atrophy. We will thus also query archived cardiac samples irradiated at the Brookhaven National Laboratory. We will also query archived cardiac samples irradiated at the Brookhaven National Laboratory involving combined X-Ray and HZE radiation, where mice with a single p53 allele deletion were irradiated. References Moffitt JA, Henry MK, Welliver KC, Jepson AJ, Garnett ER. (2013) Hindlimb unloading results in increased predisposition to cardiac arrhythmias and alters left ventricular connexin 43 expression. Am J Physiol Regul Integr Comp Physiol. 304(5):R362-73. Kwak, H.-B., W. Song,, and J.M. Lawler. (2006) Exercise-training ameliorates age-induced elevation in Bax/Bcl-2 ratio, apoptosis, and remodeling in the aging rat heart. FASEB J.
Task Progress:	Intervention with Fish Oil, Pectin: TGF-ß positive staining was elevated in the irradiated hearts. These data emphasize the importance of finding effective and safe countermeasures to mitigate radiation-induced damage and fibrosis in the heart of astronauts to address in flight and post-flight cardiovascular risk We expect Nox2 inhibition to reduce radiation-induced damage. We have been collaborating with Dr. Nancy Turner's laboratory at Texas A&M on two sets of radiation studies. Low LET gamma-ray exposure of mice will be conducted at Texas A&M overseen by Dr. John Ford's laboratory in Nuclear Engineering. Acute 0.5 Gy exposures will be used. Sacrifice of the mice and extraction of the heart and skeletal muscle will occur at 12 hours, 4 weeks, and 8 weeks following radiation treatment. Astronaut age (40-42 weeks) mice were split into (n=6/group) controls, non-irradiated with pectin + fish oil, X-ray irradiation, X-ray irradiation with pectin + fish oil. Pectin (6% by weight) and fish oil (15% by weight) will be provided in the diet as previously described (Cho et al. 2012). We have completed collecting samples and are currently analyzing irradiated hearts. In addition, the impact of HZE (28Si, 48Ti) radiation on cardiac markers of pro-oxidant and pro-fibrotic signaling is currently being examined. We postulate that intervention with fish oil and pectin will abrogate radiation-induced oxidative stress and fibrosis in the heart. Outcome Markers for Specific Aim 1 include (1) left ventricle damage, (2) oxidative stress, (3) Nox2 subunits (gp91phox, p67phox), (4) pro-fibrotic signaling (TGF-ß, p-smad 2/3, FSP-1), fibrosis (collagen I), and (5) nuclear damage. We expect that pectin and fish oil will reduce oxidative stress and boost stress response proteins (grp94, HSP70) concomitant with protection against pro-inflammatory signaling (TGF-ß, MMP-9, FSP-1, NF-kappaB). These data would demonstrate a reduction in fibrosis linked to antioxidant and anti-inflammatory properties of fish oil combined with pectin under low LET radiation in the mouse heart. Initial results indicated increased markers of oxidative stress and fibrosis in the irradiated heart. We are currently testing the ability of fish oil + pectin on inflammatory cell invasion in the heart exposed to HZE radiation. CD45 antigen was used as a marker for all leukocytes. Antibodies for CD68+ detected monocytes and macrophage. CD11c+ was our marker for all inflammatory macrophages, with iNOS a tag for M1 macrophages, while co-localization with CD206 for M2 macrophages. CD8+ antigen was used as a tag for T-cells. Data shows a trend towards higher infection of cardiac myocytes after radiation than controls. A smaller trend was noted for CD45+ cells invading cells with pectin + fish oil. Similar results were found for CD68-positive staining. These data are consistent with the hypothesis that radiation-induced damage and pathology in the heart is linked to chronic inflammation. Secondly, a nutritional intervention, fish oil + curcumin partially mitigated the effects of radiation on cardiac inflammation. Therefore, nutraceutical and potential antioxidant approaches may attenuate cardiac damage, inflammation, and fibrosis in astronauts, thus improving long-term health outcomes against cardiovascular disease. Reference Cho, Y., N.D. Turner, L.A. Davidson, R.S. Chapkin, R.J. Carroll, and J.R. Lupton. 2012. A chemoprotective fish oil/pectin diet enhances apoptosis via Bcl-2 promoter methylation in rat azoxymethane-induced carcinomas. Experimental Biology & Medicine 237:1387-1393.
Bibliography Type:	Description: (Last Updated: 06/05/2025)
Articles in Peer-reviewed Journals	Lawler JM, Hord JM, Ryan P, Holly D, Janini Gomes M, Rodriguez D, Guzzoni V, Garcia-Villatoro E, Green C, Lee Y, Little S, Garcia M, Hill L, Brooks MC, Lawler MS, Keys N, Mohajeri A, Kamal KY. "Nox2 inhibition regulates stress response and mitigates skeletal muscle fiber atrophy during simulated microgravity." Int J Mol Sci. 2021 Mar;22(6):3252. https://doi.org/10.3390/ijms22063252 ; PMID: 33806917; PMCID: PMC8005132 , Mar-2021