Fiscal Year:	FY 2020	**Task Last Updated:**	FY 05/01/2020
PI Name:	Schreurs, Ann-Sofie Ph.D.		
Project Title:	Candidate Nutritional Countermeasure to Mitigate Adverse Effects of Spaceflight		
Division Name:	Human Research		
Program/Discipline:			
Program/Discipline--Element/Subdiscipline:	HUMAN RESEARCH--Biomedical countermeasures		
Joint Agency Name:		**TechPort:**	Yes
Human Research Program Elements:	(1) **HHC**:Human Health Countermeasures		
Human Research Program Risks:	(1) **Bone Fracture**:Risk of Bone Fracture due to Spaceflight-induced Changes to Bone (2) **Cardiovascular**:Risk of Cardiovascular Adaptations Contributing to Adverse Mission Performance and Health Outcomes (3) **Osteo**:Risk Of Early Onset Osteoporosis Due To Spaceflight		
Space Biology Element:	None		
Space Biology Cross-Element Discipline:	None		
Space Biology Special Category:	None		
PI Email:	ann-sofie.schreurs@nasa.gov	**Fax:**	FY
PI Organization Type:	NASA CENTER	**Phone:**	650-604-6390
Organization Name:	NASA Ames Research Center		
PI Address 1:	Bone and Signaling Laboratory, Space Biosciences Division		
PI Address 2:	Bldg N236, Room 219		
PI Web Page:			
City:	Moffett Field	**State:**	CA
Zip Code:	94035	**Congressional District:**	18
Comments:			
Project Type:	GROUND	**Solicitation / Funding Source:**	2015-16 HERO NNJ15ZSA001N-Crew Health (FLAGSHIP, NSBRI, OMNIBUS). Appendix A-Crew Health, Appendix B-NSBRI, Appendix C-Omnibus
Start Date:	01/01/2017	**End Date:**	12/31/2019
No. of Post Docs:	0	**No. of PhD Degrees:**	0
No. of PhD Candidates:	0	**No. of Master' Degrees:**	2
No. of Master's Candidates:	0	**No. of Bachelor's Degrees:**	0
No. of Bachelor's Candidates:	1	**Monitoring Center:**	NASA JSC
Contact Monitor:	Norsk, Peter	**Contact Phone:**	
Contact Email:	Peter.norsk@nasa.gov		
Flight Program:			
Flight Assignment:	NOTE: End date changed to 12/31/2019 per L. Lewis/ARC (Ed., 7/23/19) NOTE: Period of performance changed to 1/1/2017-1/31/2019 per PI information; previously 10/1/2016-9/30/2018 (Ed., 7/15/19) NOTE: Extended to 9/30/2018 per PI; original end date was 9/30/2017 (Ed., 5/3/18)		
Key Personnel Changes/Previous PI:			
COI Name (Institution):	Tahimic, Candice Ph.D. (NASA Ames Research Center) Globus, Ruth Ph.D. (NASA Ames Research Center)		
Grant/Contract No.:	Internal Project		

Performance Goal No.:	
Performance Goal Text:	
Task Description:	In recent findings, we showed that dried plum (DP) diet conferred complete protection from the rapid bone loss induced by exposure to radiations, including gamma, protons, and High Z-High Energy (HZE) ions. Based on these very promising results on a new potential countermeasure for space radiation tissue damage, we propose to conduct additional studies and analyses, which are critical for moving the potential countermeasure to a higher countermeasure readiness level (CRL) level. We aim to test the DP diet to prevent bone loss induced by simulated spaceflight. This will be achieved by exposing mice to each factor (weightlessness and radiation) alone and combined. Furthermore, we will establish if DP protects other tissues at risk for astronauts, such as the central nervous system.
Rationale for HRP Directed Research:	
Research Impact/Earth Benefits:	Countermeasures that address the combined effects of simulated microgravity and ionizing radiation have not been investigated in bone. Both these factors are inherent to the spaceflight environment and thus, countermeasures must be investigated regarding their protective effect when both are in combination. We sought to evaluate the potentially differing effects of microgravity and ionizing radiation when controlled independently on bone and the two factors in combination. For the purpose of this study, we have used the hindlimb unloading model, in combination with exposure to total body irradiation (132Cs gamma radiation, at 2 Gy dose) as analogs of weightlessness and radiation exposure. The relatively higher dose of radiation (2 Gy) was chosen as a positive control dose to ensure bone loss in rodents to allow for testing DP as a countermeasure for bone loss. We sought to determine if the DP diet prevents simulated-microgravity induced bone loss (HU), as well as if the diet is also effective at preventing simulated spaceflight-induced bone loss (combination of HU+irradiation (IR). To address these questions, we analyzed both cancellous and cortical bone microarchitecture as well as bone quality. Additionally, we aimed to determine if the DP diet had the capacity to protect osteoprogenitors after exposure to simulated microgravity, an essential part in the healthy maintenance of the skeletal tissue.
Task Progress:	In this study, we investigated the bone protective potential of Dried Plum (DP) against independent and combined effects of simulated microgravity (Hindlimb Unloading, HU) and ionizing radiation (IR) on the microarchitecture and mechanical properties of skeletal tissue. A diet supplemented with DP prevented most of the simulated spaceflight-induced damages to both the appendicular (i.e., tibia) and axial (i.e., vertebrae) skeleton. When mice were fed the control diet, a relatively high dose (2 Gy) of low-LET (linear energy transfer) gamma radiation exclusively decreased the bone volume fraction (BV/TV) and trabecular separation (Tb.Sp) of cancellous tissue in the tibia. Based on our results, cancellous bone loss was caused by a thinning, not a decrease in the number, of existing trabeculae, which overall expanded the space between trabeculae. Hindlimb unloading (HU) for 14 days caused bone loss within both cancellous and cortical regions of the tibia and the L4 vertebrae. HU also led to reduced compressive strength of the vertebral body. The independent effects of HU and radiation (IR) remained of similar magnitude in each tissue compartment when mice were exposed to HU and IR simultaneously. Regardless of the treatment (with one exception, Ct.Th proximal tibia), consumption of the dried plum diet prevented detrimental skeletal changes. Mice fed the control diet and exposed solely to IR displayed a 20% decrement in percent bone volume and a 7% increase in the Tb.Sp of the tibia's cancellous region relative to the sham control. These parameters were unaffected by HU alone. HU groups exhibited a lesser 11% decrement in trabecular thickness (Tb.Th). When IR was combined with HU, BV/TV decreased by another 5%. These results indicate that HU and IR were not clearly additive in this experiment. Among the parameters analyzed, IR only induced changes in Tb.Th and Tb.Sp. Unlike reports by others, there were no significant changes in trabecular number (Tb.N) from any of the treatment groups relative to the sham-irradiated control group. When HU was combined with IR, Tb.Th decreased by 9%. Our results indicate bone loss was caused by a thinning, not a decrease in the number, of existing trabeculae, which expanded the space between trabeculae. The trabeculae contribute to percent bone volume and microarchitectural integrity. Since HU affects the cortical as well as cancellous tissue, we also determined the extent of cortical bone loss. The cortical region (cortical shell) exhibited a decrease in cortical thickness when mice fed the control diet were exposed to HU, either independently or in combination with IR. When mice were fed the DP diet, IR-induced cancellous bone loss was entirely prevented, consistent with the radio-protective results reported in our previous study. DP also protected the tibia when IR was combined with HU, which is a novel finding in this report. The decrease in trabecular thickness incurred by HU and HU + IR was entirely prevented by consumption of the DP diet. The DP diet also protected from HU-induced decrease in cortical thickness. Unlike the proximal tibia, the distal tibia did not show changes in cortical structure. In contrast, in the proximal tibia HU and IR together, not each treatment alone, caused a decrease in cortical thickness of the proximal tibia. This finding suggests that DP diet cannot fully prevent from all aspects of bone structural deficits induced by simulated spaceflight. In order to confirm our results, we examined another skeletal site, the vertebra, which is a representative axial bone. Under conditions of simulated weightlessness and consumption of control diet, BV/TV and Tt.Tb.Th of the vertebral body were reduced accompanied by a reduction in cortical thickness and cortical bone area. Collectively, these findings indicate an overall deterioration of bone via thinning of the vertebrae's trabeculae and cortical tissue. This deterioration of the microarchitecture directly impacted the overall strength of the tissue, as reflected in the decrease in maximum load tolerable by the vertebral body, as well as the decrease in material stiffness as measured by mechanical compression testing. Taken together, these changes in both microarchitecture and strength have the potential to lead to fracture. However, these changes were not observed when mice were exposed to IR alone. In contrast to other studies where high-LET 56Fe radiation was utilized in combination with HU, our results did not indicate an additive effect of IR on the deterioration of strength and structure of the L4 vertebrae. HU-induced damage to the vertebral body was entirely prevented when mice consumed the DP diet. Interestingly, there was a statistically significant increase in Tt.BV/TV for sham-irradiated, normally loaded (NL) mice fed the DP compared to the sham-irradiated, NL mice fed the control diet (CD). This elevated Tt.BV/TV was consistent for all treatment groups fed DP when compared to NL, CD-fed mice, suggesting a potential anabolic effect of DP. Treatments that are currently in use to mitigate the effects of mechanical unloading are not without limitations and risks. Exercise in combination with drug treatments such as bisphosphonates have shown beneficial effects in astronauts. However, in patients with osteoporosis, the use of bisphosphonates can increase the risk for atypical femoral fractures possibly due to suppressed bone turnover which may lead to cracks at the microscale and loss of mechanical strength.

Although rare, other notable side effects that have been reported to accompany bisphosphonate therapy including osteonecrosis of the jaw and atrial fibrillation. Another limitation of bisphosphonates is that they act mostly on osteoclasts. Osteoblastogenesis from flushed bone marrow cells of the femur was strongly inhibited by exposure to HU, indicating that HU directly damages osteoblast progenitors, potentially affecting in situ bone formation. Mineralization of the osteoblast cells from DP-fed mice was partially restored and taken together with the cell growth data, indicates that DP could prevent the loss of structure and strength by protecting the marrow-derived osteoprogenitors. Ex vivo osteoblastogenesis was only performed on mice exposed to HU because it has been shown previously that low-LET in vivo gamma radiation does not negatively impact osteoblasts.

Overall, the ability of DP to protect osteoblast progenitor cells from HU-induced damage, as shown in this study, holds much promise for development of next generation anti-osteoporotic drugs due to the possibility that DP to act on both osteoblasts and osteoclasts. Our current study is limited to short-duration HU (2 weeks); since astronauts on long-term space missions may require a countermeasure for bone loss throughout the entirety of a multi-year mission beyond low Earth orbit (LEO), it is important in future studies to determine if DP is protective for long-term exposure to simulated, or actual, spaceflight. Further studies also are needed to gain more insight into any potential long-term side effects of consuming a dried plum diet. Dried plum's potential as a countermeasure against both radiation- and microgravity-induced osteopenia such as loss of bone strength and structure in the tibia and vertebrae has important implications for astronauts in space as well as radiation workers, radiotherapy patients, and individuals with osteoporosis.

Bibliography Type:	Description: (Last Updated: 08/21/2020)
Articles in Peer-reviewed Journals	Steczina S, Tahimic CGT, Pendleton M, M'Saad O, Lowe M, Alwood JS, Halloran BP, Globus RK, Schreurs AS. "Dietary countermeasure mitigates simulated spaceflight-induced osteopenia in mice." Sci Rep. 2020 Apr 16;10(1):6484. https://doi.org/10.1038/s41598-020-63404-x ; PubMed PMID: 32300161; PubMed Central PMCID: PMC7162976 , Apr-2020