Fiscal Year:	FY 2013	**Task Last Updated:**	FY 11/16/2012
PI Name:	Mellor, Liliana F. Ph.D.		
Project Title:	Induction of Early Stages of Osteoarthritis After Exposure to Microgravity		
Division Name:	Human Research		
Program/Discipline:	NSBRI		
Program/Discipline--Element/Subdiscipline:	NSBRI--Musculoskeletal Alterations Team		
Joint Agency Name:		**TechPort:**	No
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) **Osteo**:Risk Of Early Onset Osteoporosis Due To Spaceflight		
Space Biology Element:	None		
Space Biology Cross-Element Discipline:	None		
Space Biology Special Category:	None		
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City:	Raleigh	**State:**	NC
Zip Code:	27695-7115	**Congressional District:**	4
Comments:	NOTE: formerly at Boise State University until fall 2013 (Ed., Jan 2014)		
Project Type:	Ground	**Solicitation / Funding Source:**	2011 NSBRI-RFA-11-01 Postdoctoral Fellowships
Start Date:	11/01/2011	**End Date:**	11/30/2013
No. of Post Docs:	1	**No. of PhD Degrees:**	0
No. of PhD Candidates:	0	**No. of Master' Degrees:**	0
No. of Master's Candidates:	0	**No. of Bachelor's Degrees:**	1
No. of Bachelor's Candidates:	3	**Monitoring Center:**	NSBRI
Contact Monitor:		**Contact Phone:**	
Contact Email:			
Flight Program:			
Flight Assignment:	NOTE: End date changed to 11/30/2013 per NSBRI (Ed., 10/24/13)		
Key Personnel Changes/Previous PI:			
COI Name (Institution):	Oxford, Julia (MENTOR/ Boise State University)		
Grant/Contract No.:	NCC 9-58-PF02601		
Performance Goal No.:			
Performance Goal Text:			

Task Description:	POSTDOCTORAL FELLOWSHIP Our goal is to investigate the effects of simulated microgravity on cartilage homeostasis, by exposing two chondrocyte cell lines to a modeled microgravity environment using a rotating wall vessel bioreactor. Cells exposed to microgravity will be compared to cells incubated under normal gravitational conditions as a control, as well as an arthritic-like cell model induced by treating chondrocytes with pro-inflammatory cytokines such as IL-1B and oncostatin M (OSM). Disruptions in cell-matrix interactions, changes in cytoskeletal morphology and gene up-regulation will be evaluated to determine changes in chondrocyte metabolism. We hypothesize that similar to bone, cartilage homeostasis can be compromised during exposure to microgravity, resulting in osteoarthritic-like conditions in astronauts after space missions. This study will give us a better insight into whether exposure to microgravity can increase the risk of developing early stages of osteoarthritis in astronauts. Due to the limited capacity of regeneration in articular cartilage, early detection and treatment are key components to prevent the advanced stages of cartilage degradation, which is the leading cause of disability in the U.S., limiting the activities of nearly 21 million adults. Cells incubated under simulated microgravity undergo morphological changes in the actin cytoskeleton. These changes were detected after only two days in simulated microgravity and retained for seven days. Cells under normal gravitation forces maintain the characteristic cortical morphology of healthy chondrocytes, while cells under simulated microgravity developed elongated fibers and adopted a more fibroblast-like morphology. Similar changes have been detected before in osteoarthritic chondrocytes, cells exposed to mechanical loading and cells undergoing de-differentiation. In addition, we detected several changes at the RNA level of genes associated with Wnt signaling pathway. Sclerostin, an inhibitor of Wnt signaling associated with bone density loss in space, was up-regulated in chondrocytes under simulated microgravity. There is not much known about the effects of sclerostin in articular cartilage, but a recent report suggested that sclerostin may protect cartilage from degradation because this up-regulation was also associated with decreased expression of several cartilage degrading genes. However, in addition to sclerostin up-regulation we also detected up-regulation of MMP-9 and down-regulation of aggrecan, both previously associated with arthritic conditions. Lastly, in a collaborative effort with Dr. Jeff Willey from Wake Forest Baptist Medical Center, we are investigating the combined effects of radiation and microgravity in articular cartilage. Radiation alone up-regulated many genes involved with OSM/IL-6 signaling pathway, suggesting that these cells are activating a pro-inflammatory reaction. The morphological cytoskeleton of irradiated cells exposed to simulated microgravity had a more drastic change than that of microgravity alone. Our goal for the next year is to look at changes at the tissue level by isolating articular cartilage explants from bovine hooves and exposing them to simulated microgravity. In addition we will continue our collaborative efforts with Dr. Willey, as well as further investigating the effects of Wnt signaling pathway in chondrocytes and simulated microgravity.
Rationale for HRP Directed Research:	
Research Impact/Earth Benefits:	It has been demonstrated that the musculoskeletal system is highly affected by radiation and microgravity during spaceflight, and there are exercise and nutrition countermeasures to prevent or minimize bone density loss and skeletal muscle atrophy after space missions. However, the effects of microgravity and radiation on articular cartilage health of the synovial joints have not been investigated, and pathological conditions such as arthritis can result in severely restricted mobility. Currently, there is no cure for arthritis due to the lack of understanding of the molecular mechanisms that trigger cartilage degradation. This is the first study to investigate the effects of radiation and simulated microgravity at the molecular level using several chondrocyte cell lines and a Rotating Wall Vessel bioreactor to simulate reduced microgravity. Our study found that chondrocytes respond to reduced gravity by changing their cytoskeletal morphology, similar to the morphological changes shown in studies using mechanical loading. We are still investigating the mechanism and mechanoreceptors responsible for re-arranging the cell morphology in response to changes in gravitational forces. Another interesting finding is the change in gene expression of molecules associated with the Wnt signaling pathway in response to simulated microgravity. The role of Wnt signaling in cartilage is well understood in embryology and development, and it has recently emerged as a critical regulator of bone and cartilage homeostasis. However, the role of Wnt signaling in arthritis is not understood, and recent studies found that sclerostin, an inhibitor of Wnt signaling, was up-regulated in mineralized cartilage and end-stage osteoarthritic samples. Sclerostin has been implicated in bone density loss in microgravity, and is now a promising therapeutic target to protect bones during space missions as well as in patients suffering from osteoporosis here on Earth. Our study is the first to detect sclerostin up-regulation in chondrocytes exposed to simulated microgravity. The mechanisms and effects of sclerostin up-regulation and changes to the Wnt signaling pathway on cartilage homeostasis will be further investigated. If Wnt signaling is in fact associated with the onset of osteoarthritis as suggested recently by a few studies, the findings from our current study may help with the understanding of the underlying mechanism involved in osteoarthritis, and can help provide a new therapeutic target for the millions of people suffering from arthritis here on Earth. Lastly, we detected the up-regulation of several genes involved in OSM/IL-6 signaling pathway in samples that received a 1Gy dose of radiation. Similar expression of these same genes have been detected in chondrocytes treated with OSM and IL-1B inflammatory cytokines, suggesting that radiation may trigger a similar reaction to that of cartilage exposed to inflammation. This is another novel finding that will require further investigation.
Task Progress:	Aims: 1: Investigate changes in gene expression after exposure to simulated microgravity. Hypothesis: Simulated microgravity will up-regulate pro-catabolic and anti-anabolic genes similar to the ones expressed after OSM and IL-1B pro-inflammatory cytokine treatments, characteristic of arthritic conditions. These changes will be assessed by RT-PCR and western blotting techniques. 2: Examine changes in cell-matrix interactions in response to simulated microgravity. Hypothesis: Exposure to simulated microgravity will have an effect on cell-matrix interactions resulting in a signaling cascade that alters the catabolic rate of chondrocytes. 3: Examine the effects of simulated microgravity on the cytoskeletal morphology of chondrocytes. Hypothesis: Chondrocytes respond to mechanical loading by changing their cytoskeletal morphology. Cells will have a similar change in morphology in response to unloading. Cytoskeletal changes will be studied by immunofluorescent staining and confocal microscopy techniques.

Bibliography Type:	Description: (Last Updated: 11/12/2020)