Fiscal Year:	FY 2013	**Task Last Updated:**	FY 05/23/2013
PI Name:	Bowles, Dawn Ph.D.		
Project Title:	Proteomic Profiling of Human Heart Tissue Exposed to Microgravity		
Division Name:	Human Research		
Program/Discipline:	HUMAN RESEARCH		
Program/Discipline--Element/Subdiscipline:	HUMAN RESEARCH--Biomedical countermeasures		
Joint Agency Name:		**TechPort:**	No
Human Research Program Elements:	(1) **HHC**:Human Health Countermeasures		
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		
PI Email:	dawn.bowles@duke.edu	**Fax:**	FY
PI Organization Type:	UNIVERSITY	**Phone:**	919-668-1947
Organization Name:	Duke University		
PI Address 1:	Department of Surgery		
PI Address 2:	Msrb1 Room 401B, DUMC 2642		
PI Web Page:			
City:	Durham	**State:**	NC
Zip Code:	27710-0001	**Congressional District:**	4
Comments:			
Project Type:	GROUND	**Solicitation / Funding Source:**	2011 Crew Health NNJ11ZSA002NA
Start Date:	07/01/2012	**End Date:**	12/31/2013
No. of Post Docs:	0	**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:**	0
No. of Bachelor's Candidates:	0	**Monitoring Center:**	NASA ARC
Contact Monitor:	Smith, Jeffrey	**Contact Phone:**	650-604-0880
Contact Email:	jeffrey.d.smith2@nasa.gov		
Flight Program:			
Flight Assignment:	NOTE: Extended to 12/31/2013 (per PI/A.Chu--NASA ARC)--Ed., 5/28/2013		
Key Personnel Changes/Previous PI:			
COI Name (Institution):	Milano, Carmelo (Duke University) Moseley, Martin (Duke University)		
Grant/Contract No.:	NNX12AK76G		
Performance Goal No.:			
Performance Goal Text:			

Task Description:	Our proposal addresses the risks of cardiac damage that could ensue during long term manned space flight. We will accomplish this by examining global protein (proteomic) changes that occur following exposure of cells and tissues to simulated microgravity, one of the stresses during manned spaceflight. In our studies we will examine the effect of microgravity on the proteome of a widely used cardiac cell type (rat neonatal cardiomyocytes) and well as on specimens of human heart tissue. The utilization of cardiac tissues from humans is a meaningful, relevant, and novel model that can directly address the NASA concerns. In addition, proteomics is a sensitive and cutting edge method to monitor protein changes on these human tissues induced by stresses that astronauts encounter during space flight. Being able to identify cardiac damage at an early stage will allow countermeasures to be administered earlier and may mitigate or reverse damage and prevent end stage heart failure. Results from this study may lead to the development of a simple blood test that could be performed on a chip to identify the current state of the cardiovascular system.
Rationale for HRP Directed Research:	
Research Impact/Earth Benefits:	In our simulated microgravity experiments we are examining the proteomics changes occurring to cardiac cells and tissues exposed to microgravity compared Earth gravity controls. Therefore, the extensive proteomics data set derived from the Earth gravity controls will be used to inform other studies that perform comparative analysis of cardiac cells exposed to other forms of stress relevant to heart injury and disease. The extensive proteomics data set can also be mined for new targets of heart disease or injury diagnosis or treatments. The data mining may enable new discoveries and products relevant to human heart disease that may be quickly translated into clinical applications. Clinical practice has the potential to be impacted and improved, for example, by the development of assays to assess cardiotoxicity of drugs, new biomarkers to assess disease progression and improvement, and new and better targeted therapeutics for cardiac disease, injury, and failure.
Task Progress:	To date we have focused on Aim 1: Aim 1. To determine the proteomic and phosphoproteomic changes which occur to rat neonatal cardiomyocytes (RNNCM) upon exposure to microgravity. RNNCM will be placed in the NASA bioreactor, also referred to as the Rotating Wall Vessel (RWV). RNNC were chosen for the first aim in this pilot as they are a readily attainable and hardy cell type which can withstand being cultured for weeks, for determining the minimum time needed for observable simulated microgravity effects via proteomics analysis. Initially we planned to evaluate proteomic changes at 1 week, 2 week, and 1 month of (+/-) exposure to simulated microgravity. Although we proposed to evaluate changes at 1 week, 2 week, and 1 month, our preliminary data (on the poster presented at the HRP Investigators' Workshop, Galveston 2013) indicated that proteomics changes were observed as soon as 12 hours post exposure to simulated microgravity. Therefore in our series of experiments for Aim1 we modified the experiment to evaluate changes at 12, 48, and 120 hours of simulated microgravity. In addition, we modified our experiments to include stable isotope labeling by amino acids in cell culture (SILAC) to evaluate changes in protein synthesis as a consequence of microgravity exposure. These experiments described above were performed three independent times. The samples were then quantified using mass spectrometry proteomics analysis by the Duke Proteomics Core Facility. Data analysis resulted in over 6,100 peptides and 848 proteins quantified, with 492 proteins having more than 1 peptide to match. There were over 1,400 peptides identified as having incorporated amino acid isotopes. The indicates that incorporation rates can be calculated for many proteins and that we will be able to evaluate protein levels change as a result in protein synthesis or degradation. We have just begun to analyze the data. The samples (microgravity and 1x gravity) are trending together with time. We plan to begin to analyze the data between time points, to determine what is differentially expressed between for example 48 hr and 12 hr, and 120 hr versus 12 hr. We plan to perform these analyses independently for the microgravity and 1x gravity controls. This type of analysis should provide us with information on the changes occurring to protein expression as a function of time. A second analysis will help confirm these findings but will specifically compare expression at 12, 48, and 120 hr between microgravity and 1x gravity. This should help us to classify quickly the proteins that are changing differently as a function of time, between 1x gravity and microgravity.
Bibliography Type:	Description: (Last Updated: 07/11/2023)
Abstracts for Journals and Proceedings	Feger BJ, Thompson JW, Moseley MA, Carnell LS, Bowles DE. "SILAC incorporation and tolerability in primary heart cells: A pilot study for microgravity-induced proteome alterations." 2013 NASA Human Research Program Investigators' Workshop, Galveston, TX, February 12-14, 2013. 2013 NASA Human Research Program Investigators' Workshop, Galveston, TX, February 12-14, 2013. , Feb-2013