Fiscal Year:	FY 2013	**Task Last Updated:**	FY 06/11/2013
PI Name:	Boyle, Richard Ph.D.		
Project Title:	Inner Ear Otoconia Response in Mice to Micro- and Hyper-gravity		
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) **Sensorimotor**:Risk of Altered Sensorimotor/Vestibular Function Impacting Critical Mission Tasks		
Space Biology Element:	None		
Space Biology Cross-Element Discipline:	None		
Space Biology Special Category:	None		
PI Email:	richard.boyle@nasa.gov	**Fax:**	FY
PI Organization Type:	NASA CENTER	**Phone:**	650-604-1099
Organization Name:	NASA Ames Research Center		
PI Address 1:	Building N239, Rm 205		
PI Address 2:	RESEARCH AST, BIOLOGICAL STUDIES		
PI Web Page:			
City:	Moffett Field	**State:**	CA
Zip Code:	94035-1000	**Congressional District:**	18
Comments:			
Project Type:	Ground	**Solicitation / Funding Source:**	2011 Crew Health NNJ11ZSA002NA
Start Date:	07/01/2012	**End Date:**	03/31/2014
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:	1	**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 3/31/2014 (original end date was 6/30/2013) per A. Chu/ARC (Ed., 6/11/13)		
Key Personnel Changes/Previous PI:			
COI Name (Institution):			
Grant/Contract No.:	Internal Project		
Performance Goal No.:			
Performance Goal Text:			
Task Description:	Does exposure to long-duration spaceflight lead to neural structural alterations and does this remodeling impact cognitive and functional performance? This knowledge gap (SM26) recognizes an inherent risk to crew health and performance due to neural structural plasticity associated with space flight. Otoconia crystals of the mammalian inner ear otolith sensory organs are critical for spatial orientation and balance. Because of their normal biomineralization and their density is related to neural sensitivity, exposure to long-duration spaceflight puts them at risk to structural remodeling. A widely considered mechanism by which the nervous system responds to a change in gravity load is a change in the weight-lending otoconia. When subjected to weightlessness, it is argued the organism counters the loss of gravity load by increasing calcium carbonate production, thereby seen as a means to increase the "system gain". Our hypothesis is: weightlessness over a significant period of time triggers a compensatory mechanism that leads to a constructive process of ion deposition and an increase of otoconia mass. Upon entry to a novel gravity environment and later return to Earth, this response is maladaptive and will have a severely negative impact on cognitive and functional performance of the crew during the mission and on health and wellness at home. Although not mutually exclusive, we also hypothesize that long-duration hypergravity exposure leads to an ablative process and loss of otoconia mass. On Earth the clinical syndrome of canalithiasis, the most common single cause of vertigo, is now clearly biomechanical in origin and occurs when otoconia or fragments from them are displaced from their normal location. Despite this significant morbidity, the potential exists for structural remodeling of otoconia by the intensity and duration of gravity loading to which the animal is exposed? To address this risk we have one specific aim, namely to specify the structural integrity of otoconia as a result of short- and long-duration exposures to altered gravity conditions. Until recently, mammalian studies were confined to space missions and ground-based centrifugation studies of relatively short duration, and as a result studies have reached mixed conclusions. The Mouse Drawer System (MDS) housed mice on the International Space Station (ISS) for 91 days, roughly 20% of the lifespan of a mouse in the wild. Preliminary results of inner ears of MDS flight mice showed a dramatic alteration of symmetry and topographical surface features of otoconia; controls were normal in appearance. Subsequent studies have acquired otoconia samples from: 1) equivalent mouse models of MDS mission to 2G centrifugation and hindlimb unloading for 91 days in two separate series; and 2) inner ear samples of mice flown on the 13-day STS-133 and -135 missions. Preliminary results of inner ears of 2G mice also showed a dramatic alteration in topographical features of otoconia, but in the opposite sense in support of our hypothesis. The proposed research is a one year ground-based study from existing tissues and addresses fundamental mechanisms of neural compensation that directly effect crew health and performance during the exploration missions and on return to Earth. We will apply scanning and transmission electron microscopy and microstructural-crystallographic techniques to evaluate the possible mechanisms of otoconia restructuring in response to gravity loading. We specifically seek to answer the following questions: are there structural changes in otoconia as a result of experimental altered gravity conditions? If so, is the change due to a constructive or destructive process? And, is the process dependent on length of exposure to altered gravity loading? It is anticipated that the study will produce both a path toward quantification of a crew health and performance risk and provide the basis for valid ground-based studies for countermeasure development.		
Rationale for HRP Directed Research:			
Research Impact/Earth Benefits:	Inner ear structures have adaptive processes to regulate their function. Because of low endolymphatic levels of calcium and carbonate ions, efficient concentrating mechanisms mediated predominantly by glycoproteins are required for crystal nucleation and growth (Thalmann I, Hughes I, Tong BD, Ornitz DM, and Thalmann R. 2006. Microscale analysis of proteins in inner ear tissues and fluids with emphasis on endolymphatic sac, otoconia, and organ of Corti. Electrophoresis 27: 1598-1608). Otoconia function is not fixed: the shell can turn over (Thalmann R, Ignatova E, Kachar B, Ornitz DM, and Thalmann I. 2001. Development and maintenance of otoconia: biochemical considerations. Ann NY Acad Sci 942: 162-178), and they slowly and progressively degenerate in aging, resulting in loss of balance and falls in elderly patients. Clinical syndrome of canalithiasis (Epley JM. 1994. Positional vertigo related to semicircular canalithiasis. Otolaryngology Head and Neck Surgery 112: 154-161), the most common single cause of vertigo, is biomechanical in origin and occurs when otoconia fragments are displaced from their normal location. Despite the significant morbidity, little is known about the structural processes involved in otoconia maintenance, and possible pathology, from long-term weightlessness.		

Task Progress:	The specific aim and project remain as originally proposed. A No Cost Extension was requested because we have finally found access to the Focused Ion Beam and Scanning Electron Microscope (dual beam) configuration needed to validate our hypothesis. Images have been acquired, and new imaging sessions are scheduled. Although delayed, we have made significant progress and are on course to successful complete the project. Specific Aim: Is the structure of inner ear otoconia remodeled by the intensity and duration of gravity loading? To answer this question we will determine the structural integrity of otoconia as a result of short- and long-duration exposure to altered gravity conditions. Mammals possess a highly conserved and elaborate gravitoinertial sensing system in the inner ear, comprised of two otolith organs, the utricle and saccule, using biomineral crystalline deposits of calcium carbonate (CaCO3) called otoconia. Otoconia formation begins during embryogenesis and is completed in early postembryonic stages. The calcium-containing part of the otoconia forms the outer layer, and it has the capacity to turn over. In weightlessness, it is argued the organism counters the loss of the gravity vector by increasing CaCO3 production and deposition onto existing otoconia, in an attempt to restore correct biomechanical interaction between movement and neural sensation. In hypergravity, it is argued the opposite occurs and the otoconia lose mass. Results and Progress The entire sample populations are now tabulated into two groups: 1) samples completely analyzed and 2) samples that will be either scanned using the standard SEM or milled using the focus ion beam/SEM instrument. It was necessary to refrain from imaging all the samples using the standard protocol of sputter-coating to the samples. This thin layer of conductive material might be inappropriate for the FIB technique, and thus samples were held in reserve. We now have images of otoconia from FIB. I am not satisfied with the results so far, and I am confident that with a slower milling technique we will be able to visualize the outer shell and inner organic core of the otoconia. This will be particularly desired for the flight otoconia where it appears a deposition has resulted from the weightlessness exposure. FIB imaging results obtained from FEI dual beam FIB/SEM show the focused ion beam can directly modify or "mill" the otoconia surface with nanometer precision. We are gaining experience in this technique and specifically we must carefully control the energy and intensity of the ion beam to create a clear cross-sectional image of the otoconia and reveal the inorganic layers and the boundary interface between the outer shell and inner organic core matrix. To conclude the study we will characterize using FIB/SEM technique the otoconia from control mice and the experimental (micro-G and hyper-G mice).
Bibliography Type:	Description: (Last Updated: 09/17/2021)
Abstracts for Journals and Proceedings	Boyle R, Varelas J. "Influence of duration and magnitude of gravity loading on mouse inner ear otoconia." 2012 NASA Human Research Program Investigators' Workshop, Houston, TX, February 14-16, 2012. 2012 NASA Human Research Program Investigators' Workshop, Houston, TX, February 14-16, 2012. , Feb-2012
Abstracts for Journals and Proceedings	Boyle R. "Inner ear otoconia response in mice to micro- and hyper-gravity." 1st Annual International Space Station (ISS) Research and Development Conference, Denver, CO, June 26-28, 2012. 1st Annual International Space Station (ISS) Research and Development Conference, Denver, CO, June 26-28, 2012. , Jun-2012
Abstracts for Journals and Proceedings	Boyle R, Popova Y, Varelas J, Kondrachuk A, Balaban P. "Influence of magnitude and time course of altered gravity on the vestibular system in fish, snails, and mice." Society for Neuroscience 2012, New Orleans, LA, October 13-17, 2012. Program#/Poster#: 574.08/HH20. Abstract available at: http://www.abstractsonline.com/Plan/ViewAbstract.aspx?sKey=5353275c-c339-4b52-b456-feb5782bb6b9&cKey=f383c367-adab-46db-83b3-9a868587ca0a&mKey=70007181-01c9-4de9-a0a2-eebfa14cd9f1 ; accessed 6/12/2013. , Oct-2012
Abstracts for Journals and Proceedings	Boyle R, Popova Y, Varelas J, Kondrachuk A, Balaban P. "Influence of magnitude and time course of altered gravity on the vestibular system in vertebrates." 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
Abstracts for Journals and Proceedings	Boyle R, Popova Y, Varelas J, Kondrachuk A, Balaban P. "Influence of magnitude and time course of altered gravity on the vestibular system in fish, snails, and mice." To be presented at the 19th IAA Humans in Space Symposium, Cologne, Germany, July 7-13, 2013. 19th IAA Humans in Space Symposium, Cologne, Germany, July 7-13, 2013. Submitted and accepted but PI's participation in the congress and presentation of this research is prevented due to the "sequestration." , Jul-2013