Task Book Report Generated on: 04/24/2024

Project Title: Contribution of the Vestibular and Sympathetic Nervous Systems to Space-Induced Bone Loss  Division Name: Human Research Program/Discipline: NSBRI  Program/Discipline: NSBRI—Musculoskektal Alterations Team  Diement/Subdiscipline: NSBRI—Musculoskektal Alterations Team  Diement/Subdiscipline: NSBRI—Musculoskektal Alterations Team  Diement/Subdiscipline: No  Human Research Program Elements: () HMC-Human Health Countermeasures  (1) Human Research Program Elements: () Hone Fracture Risk of Bone Fracture due to Spaceflight-induced Changes to Bone (2) Outer-Risk of Early Onset Osteoporosis Due To Spaceflight  None  Space Biology Cross-Element  None  Space Biology Cross-Element  None  Space Biology Special Category: None  pt Email: guillaume Frigansa/grandschill.ech  Pt Email: guillaume Frigansa/grandschill.ech  Pt Address 1: University  Pt Organization Type: University  Pt Address 2: L11 Medical Center Drive  Pt Web Page:  City: Nashville State The Control of Bone Biology  Pt Address 2: L211 Medical Center Drive  Pt Web Page:  City: Nashville State: 5  Comments:  Project Type: GROUND Solicitation Frauding 2011 NSBRI-RFA-11-01  No. of Post Doce: 1  101/2011 End Dute: 1031/2013  No. of PhD Candidates: 0  No. of PhD Candidates: 0  No. of Master's Candidates: 0  No. of Bachelor's Candidates: 0  No. of Bachelor's Candidates: 0  Contact Monitor: Contact Phone:  Contact Humil: Elepha Assignment: Key Personnel Changes/Presions Piccolated (MBNTOR/ Vanderbit Liniversity)  Performance Goul No.	Fiscal Year:	FY 2013	Task Last Undated	EV 11/16/2012
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Element/Subdiscipline:  NSBRIMuscutoskeletal Alterations Feam  Joint Agency Name:  (I) HIC-Human Health Countermeasures  (I) HIC-Human Health Countermeasures  (I) Bone Fracture-Risk of Bone Fructure due to Spaceflight-induced Changes to Bone (2) Osteo-Risk Of Early Onset Osteoporosis Due To Spaceflight induced Changes to Bone (2) Osteo-Risk Of Early Onset Osteoporosis Due To Spaceflight induced Changes to Bone (2) Osteo-Risk Of Early Onset Osteoporosis Due To Spaceflight induced Changes to Bone (2) Osteo-Risk Of Early Onset Osteoporosis Due To Spaceflight induced Changes to Bone (2) Osteo-Risk Of Early Onset Osteoporosis Due To Spaceflight induced Changes to Bone Risk Of Early Onset Osteoporosis Due To Spaceflight induced Changes to Bone Practure Risk of Bone Fracture Risk of Bone Risk Of Early Onset	Program/Discipline:	NSBRI		
Human Research Program Elements: (I) HHC-Human Health Countermeasures  Human Research Program Risks: (I) Bone Fracture Risk of Bone Fracture due to Spaceflight-induced Changes to Bone (2) Osteo-Risk of Early Onset Osteoporosis Due To Spaceflight  None  Space Biology Cross-Element: None  Space Biology Cross-Element: None  Space Biology Special Category: None  Pt Email: guillaume Evignaux@canderbit.edu Fax: FY  Pt Organization Type: UNIVERSITY Phone: 615-322-7883  Organization Name: Vanderbit University  Pt Address 1: Center for Bone Biology  Pt Address 2: 1211 Medical Center Drive  Pt Web Page:  City: Nashville State: TN  Zip Code: 37232 Congressional District: 5  Comments:  Project Type: GROUND Solicitation / Funding Source: Postocoral Fellowships  Start Date: 11/01/2011 End Date: 10/31/2013  No. of PhD Degrees: 0  No. of PhD Candidates: 0 No. of Master' Degrees: 0  No. of PhD Candidates: 0 No. of Master' Degrees: 0  No. of Master's Candidates: 0 Monitoring Center: NSBRI  Contact Monitor: Contact Phone:  Flight Assignment: Key Personnel Changes/Previous Pt:  Flight Assignment: Key Personnel Changes/Previous Pt:  COI Name (Institution): Elefteriou, Florent (MENTOR/ Vanderbit University)  Grant/Contract No.: NCC 9-58-PF02603	Program/Discipline Element/Subdiscipline:	NSBRIMusculoskeletal Alterations Team		
Human Research Program Risks:  (1) Bone Fracture-Risk of Bone Fracture due to Spaceflight -induced Changes to Bone (2) Osteor-Risk of Early Onset Ostooprosis Due To Spaceflight  Space Biology Cross-Element Discipline:  None  None  PI Email:  vullaume Evignass/divanderbilt.edu  Fax: FY  Plo Organization Type:  UNIVERSITY  Phone: 615-322-7883  Organization Name:  Vanderbilt University  Pl Address 1:  Center for Bone Biology  Pl Address 2:  1211 Medical Center Drive  Pl Web Page:  City:  Nashville  37232  Congressional District:  Sources:  Project Type:  GROUND  Solicitation / Funding Sources:  Project Type:  GROUND  Solicitation / Funding Sources:  Project Type:  Organization Page:  1 1/01/2011  End Date:  10/31/2013  No. of Phat Degrees:  No. of Phat Degrees:  No. of Phat Degrees:  No. of Phat Candidates:  O  No. of Master's Candidates:  O  Moniforing Center:  NSBRI  Contact Phone:  Contact Email:  Flight Program:  Flight Program:  Flight Program:  Flight Program:  Elefteriou, Florent (MENTOR/ Vanderbilt University)  Grant/Contract No.:  NCC 9-58-PF02603	Joint Agency Name:	1	TechPort:	No
Space Biology Element: None Space Biology Cross-Element Discipline: None Space Biology Cross-Element Discipline: None Space Biology Special Category: None PI Email: guillaume Leienaux (avanderbittedu Fax: FY PI Organization Type: UNIVERSITY Phone: 615-322-7883 Organization Name: Vanderbit University PI Organization Name: Vanderbit University PI Address 1: Center for Bone Biology PI Address 2: 1211 Medical Center Drive PI Web Page: City: Nashville State: TN Comments: Project Type: GROUND Solicitation / Funding Source: Project Type: GROUND Solicitation / Funding State Date: 110/12011 End Date: 1031/2013 No. of Post Ducs: 1 100/12011 End Date: 1031/2013 No. of PhD Degrees: 0 No. of Master's Candidates: 0 No. of Master' 0 Degrees: 0 No. of Master's Candidates: 0 Monitoring Center: NSBRI Contact Monitor: Contact Email: Flight Assignment: Key Personnel Changes/Previous PI: COI Name (Institution): Elefteriou, Florent (MENTOR/ Vanderbit University) Grant/Contract No.: NCC 9-58-PF02603	<b>Human Research Program Elements:</b>	(1) HHC:Human Health Countermeasures		
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Discipline: "None Space Biology Special Category: None PI Email: guillaume f.vignaux@vanderbilt.edu Fax: FY PO Organization Type: UNIVERSITY Phone: 615-322-7883  Organization Name: Vanderbilt University PI Organization Name: Vanderbilt University PI Address 1: Center for Bone Biology PI Address 2: 1211 Medical Center Drive PI Web Page: City: Nashville State: TN Zip Code: 37232 Congressional District: 5  Comments: Project Type: GROUND Solicitation / Funding 2011 NSBRI-RFA-11-01 Source: Postdoctoral Fellowships Start Date: 11/01/2011 End Date: 10/31/2013  No. of Post Does: 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: 0 Monitoring Center: NSBRI Contact Monitor: Contact Phone: Flight Program: Flight Program: Flight Assignment: Key Personnel Changes/Previous PI: COI Name (Institution): Elefleriou, Florent (MENTOR/Vanderbilt University) Grant/Contract No.: NCC 9-58-PF02603 Performance Goal No.:	Space Biology Element:	None		
Pl Email:   guillaums Evignaux@vanderbilt.edu   Fax: FY     Pl Organization Type:   UNIVERSITY   Phone: 615-322-7883     Organization Name:   Vanderbilt University     Pl Address 1:   Center for Bone Biology     Pl Address 2:   1211 Medical Center Drive     Pl Web Page:   State: TN     Zip Code:   37232   Congressional District: 5     Comments:   Comments:     Project Type:   GROUND   Solicitation   Funding Source:     Project Type:   GROUND   Solicitation   Funding Source:     Project Type:   1/01/2011   End Date:   10/31/2013     No. of Post Docs:   No. of PhD Degrees: 0     No. of PhD Candidates:   0   No. of PhD Degrees: 0     No. of Master's Candidates:   0   Monitoring Center:   NSBRI     No. of Master's Candidates:   0   Monitoring Center:   NSBRI     Contact Monitor:   Contact Phone:	Space Biology Cross-Element Discipline:	None		
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## POSTDOCTORAL FELLOWSHIP

a manner that is more complicated than anticipated.

Our main hypothesis is that the vestibular system participates to the maintenance of bone mineral density on Earth and its dysfunction under microgravity may contribute to the bone loss associated with space travel. Our preliminary findings have provided evidence to support our hypothesis. Bilateral vestibular lesion (VBX) using sodium arsanilate injections in rats led to significant bone loss associated to a decrease in osteoblasts number.

Aims of this project are 1) to determine if VBX causes bone loss by activation of the sympathetic nervous system in our VBX model using beta-blocker-treated mice and mice lacking the beta-2 adrenergic receptors globally or specifically in osteoblasts; 2) to analyze the bone phenotype of mice devoid of vestibular gravity sensor (Het-/- mice); and 3) to test nitric oxide involvement in vestibular-related bone loss using a vestibular hair cells specific KO for Sod3. Our study may uncover a new pathway of bone regulation, a novel approach for the treatment of low bone mass diseases on Earth, and novel countermeasures to reduce risk of bone fracture in microgravity.

During this first year of research we confirmed our previous results obtained in rats in a mouse model of vestibular lesion. Indeed 2-month old mice displayed reduced bone mineral density in femurs 1 month after VBX, as observed in rats. This first result is important as it allows us to use genetically-modified mutant mice in future studies. We also investigated the SNS involvement in VBX-induced bone loss in mice. We found that daily propranolol treatment prevented VBX-induced bone loss, as it did previously in rats, and beta-2 adrenergic receptors KO mice femurs were not sensitive to VBX. Both results support the hypothesis of a SNS involvement in our model. Finally, no bone change was observed in 3-month old Het-/- mice (Nox3het/J) (mice lacking otoliths). Nox3 encodes a NADPH oxidase which is involved in the down-regulation of nitric oxide (NO) availability. Knowing that NO level in vestibular hair cells modulates vestibular signals and that it is an important neuromediator/neuromodulator of the vestibular response, we

Aim 3 of this project will help teasing apart the mechanism involved. We are currently breeding conditional beta-2 adrenergic receptors KO mice lacking this receptor specifically on osteoblasts in order to confirm, genetically and without the possible complications of developmental phenotypes, the SNS involvement in VBX-induced bone loss. We will also start aim 3 of our project which, in addition to results of aim 2, will help us to better understand the role of NO in vestibular response generation in hair cells.

hypothesize that the combination of lack of otoliths and impaired neurotransmission possibly impact bone remodeling in

## Rationale for HRP Directed Research:

**Task Description:** 

## **Research Impact/Earth Benefits:**

The goal of this project is to address the existence of a sensory and neuronal-based mechanism by which microgravity conditions provoke bone loss, using genetic and pharmacological in vivo approaches, with the long-term goal to propose novel and more targeted pharmacological avenues to prevent microgravity-associated bone loss. Our results, beyond contributing to our understanding of the mechanisms whereby bone homeostasis is controlled, have potential important clinical implications. One is that beta-2 AR pharmacological blockade may be able to counteract the bone loss associated with unloading conditions during bed rest or long-term space travel. Another implication is that patients with vestibular pathologies, especially bilateral dysfunctions, may present with low bone mineral density and be at higher risk for fracture than the general population. This may also be relevant to the association between vestibular dysfunctions and bone loss observed in aging individuals. Therefore, one could also speculate that the progression of bone loss during aging could be accelerated upon vestibular dysfunction. All these implications remain at the present time speculative but warrant further experimental and clinical investigations.

SPECIFIC AIM 1: Determine if vestibular lesion causes bone loss by activation of the sympathetic nervous system. The

first step of this study consisted in evaluating the effect of a bilateral vestibular lesion (VBX) on femoral bone mass. Using micro-ct analyses on 2-month old mice, we demonstrated that VBX induces a significant decrease in femoral bone mass one month after the lesions. Histomorphometric analyses are ongoing. The second step was to determine whether the sympathetic nervous system (SNS) mediates this VBX-induced bone loss. Using 2-month old beta-2 adrenergic receptors KO mice, we detected no bone change in BV/TV 1 month after VBX. Moreover, daily propranolol treatment had no effect on WT-sham mice but completely blunted VBX-induced bone loss in WT-VBX mice. Taken together, these results suggest that the SNS might be involved in VBX-induced bone loss. We are currently breeding a conditional beta-2 adrenergic receptors KO model lacking these receptors specifically on osteoblasts. We thus will be able to make the distinction between the central and peripheric SNS involvement in VBX-induced bone loss. SPECIFIC AIM 2: Analyze the bone phenotype of mice devoid of vestibular gravity sensor. We used 3-month old Het-/- mice (Nox3het/J), lacking otoliths (gravity sensors), in order to mimic the decrease in vestibular stimulation in space. Micro-CT analyses revealed no bone changes in these mice compared to WT. Because of the lack of otoliths, we assumed that these mice should present a decrease in vestibular inputs and bone loss. However, Nox3 encodes a NADPH oxidase in hair cells which is involved in the downregulation of nitric oxide (NO) availability. Knowing that NO level in vestibular hair cells modulates the vestibular signals and that it is an important neuromediator/neuromodulator of the vestibular response, a constitutive high NO level in hair cells might explain our

SPECIFIC AIM 3: Test nitric oxide involvement in vestibular-related bone loss The Sod3flox/flox mice are currently at early stages of breeding. Preliminary experiments to define the titer of cre-adenovirus needed to inactivate Sod3 specifically in the inner ear are ongoing, using Rosa26 reporter mice. Successful vestibular histological sections have been obtained, which will be critical to measure the extent and specificity of cre-recombination in this model.

## **Bibliography Type:**

Description: (Last Updated: 04/12/2016)

results in this model (aim 3 will help to clarify this).

Task Progress: