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14. ABSTRACT

The goal of this GWIRP project is to elucidate the mechanisms that subserve the brain function impairments in Gulf War Veterans' Illness with headache, with chronic headaches and body muscle and joint pain with and without comorbid depression through exploration of different features of brain function and connectivity whose impairment characterize widely prevalent chronic pain conditions. In pursuit of these goals, we acquired and analyzed data from 5 healthy controls. We also analyzed the data of all the GWVI-HAP veterans collected in the linked clinical trials so far.

15. SUBJECT TERMS
None listed.

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INTRODUCTION

An estimated 25% to 32% of 1991 Gulf War veterans experience multi-symptom conditions not explained by stress or psychiatric illness (1). Affecting up to 200,000 veterans deployed to Iraq, Kuwait, and Saudi Arabia during the 1991 Persian Gulf War, this poorly understood chronic medical condition referred to as Gulf War Illness (GWI), comprises a variety of behavioral and neurological symptoms and complaints including fatigue, generalized neuropathic pain, memory and concentration deficits, vestibular disturbances, and depression. Chronic pain is an especially common complaint in GWVI (2-5). Among the neurologic symptoms of brain impairment GWVI-related chronic headaches and body muscle and joint pain conditions (GWVI-HAP) are the most debilitating, affecting around 64% of the GWVI veterans (2, 6). Further, depression carries a very high co-morbid rate (>50%) in patients with chronic pain, including GWVI-HAP. These co-morbid pain and depressive symptoms are often associated with other neuropsychological dysfunction in attention, memory and systemic symptoms, which cast a profound negative impact on patients' quality of life (4, 7-10). The objective/goal of this project titled, “Exploring Mechanisms underlying Dysregulation of Central Nervous System in GWVI-HAP” is to discover the brain function mechanisms underlying GWVI-HAP with and without comorbid moderate/severe depression. This search for mechanisms of GWVI-HAP was to be accomplished by mapping the whole-brain functional connectivity (FC) network architecture of GWVI extracted from resting state functional magnetic resonance imaging (rsfMRI) data with different advanced techniques.

KEYWORDS: Gulf War Veterans Illness; GWVI-related chronic headaches and body muscle and joint pain; depression; mechanisms; resting state fMRI; functional connectivity; brain function networks advanced network analysis.

ACCOMPLISHMENTS

Major goals of the project in Quarters 1-3

According to the approved **Statement of Work** for the aims and goals for of this GWIRP project Exploring Brain Mechanisms Underlying Gulf War Illness through Advanced Network Analysis are (see also Table 1; which lists the Aims and Tasks by Quarter):

Specific Aim 1: To extract brain function mechanisms underlying GWVI-HAP by examining functional connectivity in different *a priori* postulated functional network models of chronic pain (Table 1).

Specific Aim 2: To comprehensively examine brain resting state fMRI networks with independent components analysis, in order to explore brain function mechanisms underlying GWVI-HAP

Specific Aim 3: To perform classification of the subjects into GWVI-HAPnodep, GWVI-HAPdep or HC based on unique features (i.e., *connectomics signatures)* of their functional connectome, and to elucidate brain mechanisms subserving GWVI-HAPnodep and GWVI-HAPdep, based on the *connectomics signatures*.

Specific Aim 4: To examine alterations in rsfMRI signal complexity across the brain

Table 1: ***Project Timelines:*** Project task schedules by quarter (Q) are tabulated below

Tasks	Q1	Q2	Q3	Q4
Staff Recruitment	X			
fMRI Acquisition from HCs: Expected Complete			3	5
fMRI Acquisition from GWVI-HAP: Expected Complete	16*	8	8	8
rsfMRI Preprocessing		X	X	X
Specific Aim1		X	X	X
Specific Aim2		X	X	X

Specific Aim3		X	X	X
Specific Aim4		X	X	X
Dissemination of Results				

Staff Recruitment: We were able to recruit the two Graduate Research Assistants who will be working on project. Hence, staff recruitment is complete.

Acquisition of MRI data from GWVI-HAP veterans: Veterans with GWVI-HAP with and without comorbid depression are being conducted in a GWIRP Clinical trial GW180150 and VA CS R&D Merit Award (CX001986-01) both led by Dr Albert Leung. Dr. Anna Woodbury leads the Atlanta VAHCS arm of these trials. This study (GW200035) will analyze de-identified data from these projects. We were able to complete analysis of MRI data from 26 GWVI veterans whose data has been collected so far in the two above-mentioned clinical trials. These clinical trials are running behind schedule on subject recruitment. They have only collected 26 GWVI veterans' data until now. This is short of the target of 40 as per our Statement of Work.

Acquisition of MRI data from Healthy Controls: We were able to acquire and analyze 5 healthy control subjects. This is short of the target of 8 as per our Statement of Work. We had some difficulty recruiting subjects for this study, but this has been solved and we were able to obtain data from 5 healthy controls in just one month. Hence, we are certain of meeting our deadlines by the next reporting period.

Preprocessing and Parcellation of rsfMRI data: Resting state fMRI data from all subjects was preprocessed according to well-established pipelines (11, 12). The rsfMRI time-series were corrected for magnetic susceptibility induced image distortions, temporally shifted to account for differences in slice acquisition times, 3D volume registered to a base volume to account for global rigid motion; co-registered to the T1-weighted high-resolution anatomic scan using the well-established affine boundary based registration algorithm (13); spatially normalized to the

MNI152 template; and resampled to 2mm X 2mm X 2mm voxel resolution. The spatially normalized time-series data were detrended of temporally and spatially structured artifacts caused by motion, draining veins, ventricular and white-matter specific signals, susceptibility artifacts, and physiological noise with the sophisticated ICA-based FIX technique (14). The resultant time-series were detrended of low frequency drifts (with a 0.01 Hz cutoff high-pass filter), as well as well as the six rigid-body motion parameters, as well as deep cerebral white matter signals, and spatially smoothed with an isotropic Gaussian filter (FWHM = 5 mm). The pre-processed fMRI time-series were parcellated based on the well-established 276-ROI Braintome atlas (15), for Specific Aim 3. We were able to complete this task on the 26 GWVIs and 5 HCs collected so far.

Performing Seed Based Functional Connectivity Analysis (SA 1): We performed seed-based cross-correlation analysis (SB-CCA) employing seed ROIs of all brain areas in frontal, striatal, insular, limbic, and somatosensory regions implicated in processing, perception and limbic effects of pain. We were able to complete this task on the 26 GWVIs and 5 HCs collected so far.

Performing Group ICA Based Analysis of Resting State Networks (SA2):

The pre-processing (as described above) for conducting Group ICA (GICA) has been completed on 26 GWVIs and 5 HCs collected so far. GICA requires an adequate sample size in all three groups to provide meaningful reproducible results, which is not targeted to be achieved till Q6.

SVM-based Classification of subjects (SA3):

The pre-processing (as described above) for conducting support vector machine (SVM)-based classification has been completed on the 26 GWVIs and 5 HCs collected so far. SVM classification requires large sample sizes in all three groups to provide meaningful reproducible

results, which is not targeted to be achieved till Q8. Hence, it is not feasible to run SVM classification yet on the data collected.

Performing Multi-Scale Entropy Analysis (SA4):

We have completed the multi-scale entropy-based fMRI complexity analysis on the 26 GWVIs and 5 HCs collected so far. Sample entropy (SE) was calculated at 10 temporal scales to obtain multi-scale entropy (MSE). The window-length *m* for SE calculations was set to 2, and the distance threshold *r* set to 0.3 based on well-established methods (16, 17). ROI-averaged MSE was also obtained from the 276 Braintome ROIs.

Significant results or key outcomes: Nothing to Report.

Other Achievements: Nothing to Report

Opportunities for training and professional development: Not applicable to this grant

Table 2: ***Project Timelines:*** Project task schedules by quarter (Q) are tabulated below

Tasks	Q1	Q2	Q3	Q4	Q5	Q6	Q7	Q8	Q9	Q10	Q11	Q12
Staff Recruitment	X											
fMRI Acquisition from HCs: Expected Complete			3	5	5	5	5	5	5	5	2	
fMRI Acquisition from GWVI-HAP: Expected Complete	16*	8	8	8	8	8	8	8	8	8	8	8
rsfMRI Preprocessing		X	X	X	X	X	X	X	X	X	X	
Specific Aim1		X	X	X	X	X	X	X	X	X	X	X
Specific Aim2		X	X	X	X	X	X	X	X	X	X	X
Specific Aim3		X	X	X	X	X	X	X	X	X	X	X
Specific Aim4		X	X	X	X	X	X	X	X	X	X	X
Dissemination of Results							X	X			X	X

IMPACT

Impact on the development of the principal discipline(s) of the project: Nothing to report

Impact on other disciplines: Nothing to report

Impact on technology transfer: Nothing to report

Impact on society beyond science and technology: Nothing to report

CHANGES/PROBLEMS

Recruitment of GWVI veterans in the clinical trials (GWIRP Clinical trial GW180150 and VA CS R&D Merit Award (CX001986-01)) whose data this project depends on has been behind schedule. The issues and potential solutions and improvements pertaining to those trials will be addressed by the corresponding PIs in their reports. Of critical importance to this project the shortfalls in recruitments mentioned above have resulted in our analyzing only 26 of the 40 GWVI datasets targeted at the end of Q4.

Recruitment of human control subjects in the age-group needed for this project was a bit challenging. Hence, we are short of our target. However, we have found a lot of success in attracting human control subjects after we very recently put an advertisement in a newspaper targeting seniors. For instance, we were able to acquire and analyze data from 5 subjects in just one month. Hence, we are confident of meeting our goals by the next reporting period.

PRODUCTS

Nothing to report. Dissemination of Results is not scheduled to occur till Year 2.

PARTICIPANTS & OTHER COLLABORATING ORGANIZATIONS

Emory University Personnel

Name	**Kaundinya Gopinath, PhD.**

Project Role	Principal Investigator (PI) of the DoD award
Research Identifier	ERA Commons: KGOPIN
Nearest person month worked:	2.64 calendar months
Contribution to Project:	Dr. Gopinath took part in all the tasks completed. He supervised all work on the project. He shared data analysis responsibilities with the Graduate Research Assistants Gabriell Champion, and Guangming Yang. He also designed data analysis methodology adopted in the project. In this he received constructive input from the co-investigators (co-Is) Drs Ying Guo, Keith McGregor and Albert Leung. He shared administrative tasks pertaining to IRB-related matters with the graduate student Gabriell Champion.

Name	**Ying Guo, PhD.**
Project Role	Co-Investigator
Research Identifier	ERA Commons: YGUO76
Nearest person month worked:	1.2 calendar months
Contribution to Project:	Dr. Guo provided constructive input into design and execution of data analysis methodology adopted in the project.

Other collaborating Organization 1:

Organization 1 Name: Veterans Medical Research Foundation (VMRF)

Location of Organization 1: Address: 3350 La Jolla Village Dr. (151A), San Diego, Ca. 92161

Partner's contribution to the project: Collaboration through Emory-VMRF sub-contract under the DoD award for this project

VMRF Sub-Contract Personnel

Name	**Albert Leung, MD.**

Project Role	Co-Investigator
Research Identifier	ERA Commons: AYLEUNG
Nearest person month worked:	1.2 calendar months
Contribution to Project:	Dr. Leung is the main PI of the clinical trials whose data is being examined in this project. He provided constructive input into design of project methodology based on his expertise in the field of GWVI rehabilitation therapy.

Other collaborating Organization 2:

Organization 2 Name: Department of Psychology, Georgia State University

Location of Organization 2: Address: 140 Decatur St SE, Atlanta, GA 30303

Partner's contribution to the project: Collaboration through Emory-GSU sub-contract under the DoD award for this project

GSU Sub-Contract Personnel

Name	**Gabriell Champion, BS.**
Project Role	Graduate Student
Research Identifier	
Nearest person month worked:	1.2 calendar months
Contribution to Project:	Ms. Champion shared data analysis responsibilities under the supervision of Dr. Gopinath. She also performed administrative research coordination tasks including creating IRB documentation under Dr. Gopinath's guidance

Other collaborating Organization 3:

Organization 3 Name: Center for Visual and Neurocognitive Rehabilitation, Atlanta VAHCS

Location of Organization 3: Address: 1670, Clairmont Road, Decatur, GA 30033

Partner's contribution to the project: Collaboration through Emory- Atlanta VAHCS Sub-Contract

Name	**Keith McGregor, PhD.**
Project Role	Co-Investigator
Research Identifier	ERA Commons: KMCGREGOR
Nearest person month worked:	1.2 calendar months (in-kind; no salary support)
Contribution to Project:	Dr McGregor provided constructive input into design of project methodology based on his expertise in cognitive neuroscience. Dr McGregor has shifted to another university, but he still maintains courtesy appointment in Atlanta VAHCS and is able to continue participating in the project without beak

APPENDIX 1: REFERENCES

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