AWARD NUMBER:
W81XWH-17-1-0334

TITLE:

The Use of B-Cell Depletion Therapy (BCDT) in Gulf War Illness: A Phase 1/2 Study

PRINCIPAL INVESTIGATOR: Nancy Klimas

CONTRACTING ORGANIZATION: Nova Southeastern University

REPORT DATE: Oct 2019

TYPE OF REPORT: Annual

PREPARED FOR: U.S. Army Medical Research and Materiel Command
Fort Detrick, Maryland 21702-5012

REPORT DOCUMENTATION PAGE

Form Approved
OMB No. 0704-0188

1. REPORT DATE	2. REPORT TYPE	3. DATES COVERED
Oct 2019	Annual	**09/30/2018 - 09/29/2019**

4. TITLE AND SUBTITLE

The Use of B-Cell Depletion Therapy (BCDT) in Gulf War Illness:
A Phase 1/2 Study

5a. CONTRACT NUMBER
W81XWH-17-1-0334

5b. GRANT NUMBER

5c. PROGRAM ELEMENT NUMBER

5d. PROJECT NUMBER

5e. TASK NUMBER

5f. WORK UNIT NUMBER

6. AUTHOR(S)

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7. PERFORMING ORGANIZATION NAME(S) AND ADDRESS(ES)

Nova Southeastern University, 3301 College Avenue, Fort Lauderdale, FL 33314

Duke University
308 Research Drive, Durham, NC 27710

8. PERFORMING ORGANIZATION REPORT

9. SPONSORING / MONITORING AGENCY NAME(S) AND ADDRESS(ES)

U.S. Army Medical Research and Development Command
Fort Detrick, Maryland 21702-5012

10. SPONSOR/MONITOR'S ACRONYM(S)

11. NUMBER(S)

12. DISTRIBUTION / AVAILABILITY STATEMENT

Approved for Public Release, Distribution Unlimited

13. SUPPLEMENTARY NOTES

14. ABSTRACT

Gulf War Illness (GWI) (also known as Gulf War Syndrome) is a chronic multi-symptom illness that has been implicated in as many as one-third of the 700,000 U.S. troops deployed to the Middle East during the 1990-1991 Gulf War. Despite the time, expense, and effort, the cause of GWI remains relatively unknown and treatments have been targeted at improving symptoms. Veterans with GWI are still plagued by multiple symptoms including problems with fatigue, headaches, joint and muscle pain, gastrointestinal and sleep disturbances, neurologic and neuropsychological symptoms, respiratory issues, and cardiovascular problems. These symptoms are burdensome, impacting the patient's ability to work and care for themselves and loved ones, and impacting activities of daily living as well as quality of life.

While treatment remains focused on symptom improvement, there have been significant advances in gaining insight into potential biologic mechanisms that cause persistence of this disabling illness. Biomarkers have been discovered that may play a role in the onset and progression of the disease, such as markers of inflammation and immune dysfunction, an immune signature that is similar to that seen in autoantibody mediated illnesses such as rheumatoid arthritis, psoriatic arthritis, and myasthenia gravis. Specifically, the immune signature would favor the production of autoantibodies, a theory Dr. Abou-Donia pursued in animal models of GWI, and most recently has demonstrated in the serum of GWI subjects. Our current research efforts using a computational biology model have identified immune function, specifically autoantibody production, as a reasonable GWI target.

Autoantibody-mediated inflammatory illnesses have been treated effectively with B-cell depleting therapies, such as rituximab. Further, there is a closely related illness with a similar immune signature, myalgic encephalomyelitis/chronic fatigue syndrome (ME/CFS), with two Phase 2 clinical trials that have evaluated the safety and effectiveness of rituximab and have shown an improvement in 65% of those patients treated with rituximab, often with marked and sustained improvement. There have been case reports of sustained normal health for more than 4 years. This is intriguing as in ME/CFS there is no defined autoantibody, yet the immune signature that led to these studies is very similar to GWI.

Our strategy is to engage a team of highly qualified researchers to measure the clinical and biologic response to a B-cell depleting therapy (rituximab), which temporarily reduces or removes the B-cells from circulation; B-cells are responsible for antibody production, the treatment targets autoantibody production. This in turn can potentially reduce or even completely eradicate future formation of autoantibodies and reset the underlying mechanisms of disease to improve symptoms and reset homeostasis, as predicted by our computational models. The use of a B-cell depleting therapy such as rituximab not only may prove effective in treating patients but also prove that the autoantibodies seen in GWI are indeed mediators of illness persistence. If proved effective in this study, a Phase 3 study would be justified, the results of which could change the standard of care for GWI, targeting the underlying cause of disease and not just treating symptoms tied to disease persistence.

In this poorly understood disease with minimal treatment approaches, this study will provide an understanding of disease onset and progression and provide a targeted therapy for at least a subgroup of patients with GWI and drastically change the dynamic of treatment. This not only sets to determine an underlying cause of disease but change the course of treatment by moving from amelioration of symptoms to resetting immune function and memory and removing the cells producing damaging autoantibodies.

15. SUBJECT TERMS

16. SECURITY CLASSIFICATION OF:			**17. LIMITATION OF ABSTRACT**	**18. NUMBER OF PAGES**	**19a. NAME OF RESPONSIBLE PERSON**
a. REPORT	**b. ABSTRACT**	**c. THIS PAGE**			**19b. TELEPHONE NUMBER** *(include area code)*
Unclassified			Unclassified		

Standard Form 298 (Rev. 8-98)
Prescribed by ANSI Std. Z39.18

TABLE OF CONTENTS

1. **INTRODUCTION:** *Narrative that briefly (one paragraph) describes the subject, purpose and scope of the research.*

In this poorly understood disease with minimal treatment approaches, this study will provide an understanding of disease onset and progression and provide a targeted therapy for at least a subgroup of patients with GWI and drastically change the dynamic of treatment. This not only sets to determine an underlying cause of disease but change the course of treatment by moving from amelioration of symptoms to resetting immune function and memory and removing the cells producing damaging autoantibodies.

2. **KEYWORDS:** *Provide a brief list of keywords (limit to 20 words).*

Gulf War Illness, Clinical Trials, Rituximab, autoantibodies, cytokines

3. **ACCOMPLISHMENTS:** *The PI is reminded that the recipient organization is required to obtain prior written approval from the awarding agency grants official whenever there are significant changes in the project or its direction.*

What were the major goals of the project? What was accomplished under these goals?

	Timeline	**% complete**
Major Task 1: Perform a randomized phase I/II study comparing rituximab to placebo after 2 infusions 2 weeks apart, then monitored for 9 months to assess safety, efficacy and biomarker response to therapy	**Months**	
Subtask 1: Prepare Regulatory Documents and Research Protocol for Study		
Refine eligibility criteria, exclusion criteria, screening protocol	1-3	100%
Finalize consent form & human subjects' protocol	12	100%
IRB protocol submission NSU and Miami VAMC (to permit recruitment from Miami VAMC)	1-24	100%
Military 2nd level IRB review (ORP/HRPO)	12-24	100%
Submit amendments, adverse events and protocol	As needed	
Submit annual IRB report for continuing review	As needed	
Milestone Achieved: Local IRB (NSU) and HRPO approval	2-24	100%
Subtask 2: preparation for initiation of clinical trial (staff/space/platform)		

Study Staff definition of duties, cross training, Coordinate with study staff to customize web based assessment platform (REDCap) and research pharmacy a flow chart for all study steps, web data collection and database requirements	12-24	90%
Coordinate with study staff, mock run of virtual subject through all time points and review platform for HIPAA, permissions, and data retrieval issues	24-32	90%
Milestone Achieved: Research staff trained, platform tested, 1st participant consented and study begins	30-32	80%
Subtask 3: Initiation of randomized control clinical trial		
Recruitment of subjects, screening, informed consent process	24-36	20%
Completion of last subject last assessment. Monitor and report adverse events to IRB, Data monitoring board chair (*continuous per GCP, formal report quarterly)*	36	
Milestone achieved: Clinical trial underway, meeting recruitment goals *Milestone achieved last subject completes intervention and final assessment*	6-36 36	
Subtask 4: Biomarkers/lab studies		
Review established protocols and establish accession, barcoding and sample flow. Coordinate with study coordinator and lab staff on scheduling, shipping protocol for Duke samples	1-3	20%
Determine batch assays schedule, data entry plan, work with lab information system for direct data entry to REDCap platform. Maintain quality control for all assays review with lab weekly.	1-6	50%
Run assays as described in protocol, maintaining quality and timely data entry. Stats core to run monthly data status for all investigators	7-34	0%
Milestone achieved: lab information system platform link to REDCap *Milestone achieved completion of all assays with data entered into system*	6 34	80% 0%

Specific Aim 2: To perform biomarker studies of immune function, activation and cytokine expression to inform the ongoing computational modeling program and to assess impact of intervention on immune parameters.

Major Task 2: To perform biomarker studies of immune function, activation and cytokine expression to inform the ongoing computational modeling program and to assess impact of intervention on immune parameters.

Subtask 1: Computational modeling

Apply previously developed computational modeling to before/after intervention comparisons	12-36	0%
Adapt computational modeling platform for intervention before/after analyses	1-12	0%
Perform dynamic modeling utilizing group A/B designation for interim analyses	24	0%
Break blind and completion of final analyses. Final publications and translation plan	34-36	0%
Milestone achieved: Interim analyses data used in modeling data and its results	34-36	0%

Study specific aim 3: To determine the levels of autoantibodies against neuronal glial proteins as follows before administering rituximab and 6 weeks, 3 months, 6 months and 9months after initial administration of rituximab or placebo to allow an initial evaluation of the clinical condition and symptom complex of GWI veterans and the levels of autoantibodies and changes in antibody level against neural proteins.

Major Task 3: To determine the levels of autoantibodies against neuronal glial proteins as follows before administering rituximab and 6 weeks, 3 months, 6 months and 9 months after initial administration of rituximab or placebo. This will allow an initial evaluation of the clinical condition and symptom complex of veterans with GWI and the levels of autoantibodies and changes in antibody level against neural proteins.

Subtask 1: Measurement of the levels of autoantibodies against neuronal glial proteins

Autoantibody assays Measurement of circulating neural peptides	12-36	10%
Analysis correlating findings with treatment group and symptom cluster	34-36	0%
Milestone achieved: Co-author manuscript on neural autoantibodies status in GWI	24-36	0%

What opportunities for training and professional development has the project provided?
If the project was not intended to provide training and professional development opportunities or there is nothing significant to report during this reporting period, state "Nothing to Report."

Nothing to report

How were the results disseminated to communities of interest?
If there is nothing significant to report during this reporting period, state "Nothing to Report."

Nothing to report

What do you plan to do during the next reporting period to accomplish the goals?
If this is the final report, state "Nothing to Report."

Describe briefly what you plan to do during the next reporting period to accomplish the goals and objectives.

The IRB and HRPO approvals are complete, the study has been submitted as active to clincialtrials.gov.

With these approvals were have been able to initiate recruitment as follows:

The Sullivan study on CNS autoantibodies is an excellent source of potentially study subjects, the Miami site provided 1/3 of the cohort under study. The subjects in that study were pulled from the Sullivan GWIC cohort, all are consented for permission to contact about future study opportunities. Dr Abou Donia has run these samples in a blinded fashion, our co-investigator Kim Sullivan has the results and holds the blind, and the permission to re-contact the study eligible subjects.
In addition to this population, we also have the local gulf war illness veterans, both the population cared for through the community care contract with the VA at Nova Southeastern INIM clinic is and also the VA GWI clinic directed by Dr Klimas. Flyers and face to face recruitment is underway at these two sites.

These subjects recruited newly from the community or from the clinics will require a rapid turnover of the screening labs, including the autoantibody studies performed at the Duke lab of Dr Abou Donia. Dr Donia has developed more efficient testing strategies that assures a rapid (2 week) turnaround.

From the initial Miami based Sullivan cohort 20 veterans with GWI have completed autoantibody panels, all 20 had evidence of CNS autoantibodies and are study eligible at the time of their participation in the Sullivan GWIC. We received these results in the past 2 weeks, and are in the process of scheduling study willing subjects for repeat screening and enrollment at this time.

In addition we have 4 subjects from the GWI clinic is at NSU that have expressed interest and are being screened for study eligibility at this time.

It is clear the study will require a full time coordinator (we had used a part time coordinator, Ms. Renesca ARNP) in the period needed to develop the platform and the procedures) the coordinator position of this study was posted and we have interviewed 5 candidates, the position will be filled in the next 2 weeks. While we complete the training of the new coordinator we will proceed with recruitment and screening.

In order to avoid recruiting from the same pool of subjects we have studied extensively in earlier studies, we launched an independent effort to register veterans with gulf war illness using a recruitment tool that is developing a registry of study interested veterans nationally. This IRB approved on line program (not funded through a federal mechanism) is allowing our team to roll out an aggressive social media effort asking the veteran advocates to help us help them by making recruitment for studies more widely available. This tool will register veterans and ask that they take a short survey of symptoms and severity, and permits the team to engage with the interested veterans for education and research purposes. The surveys with HPI are housed in the HIPAA compliant server at NSU and will be a resource for all of the GWI research community. The beta tests were completed in June and the social media launch has begun.

4. **IMPACT:** D*escribe distinctive contributions, major accomplishments, innovations, successes, or any change in practice or behavior that has come about as a result of the project relative to:*

What was the impact on the development of the principal discipline(s) of the project?
If there is nothing significant to report during this reporting period, state "Nothing to Report."

Describe how findings, results, techniques that were developed or extended, or other products from the project made an impact or are likely to make an impact on the base of knowledge, theory, and research in the principal disciplinary field(s) of the project. Summarize using language that an intelligent lay audience can understand (Scientific American style).

Nothing to report

What was the impact on other disciplines?
If there is nothing significant to report during this reporting period, state "Nothing to Report."

Describe how the findings, results, or techniques that were developed or improved, or other products from the project made an impact or are likely to make an impact on other disciplines.

Nothing to report

What was the impact on technology transfer?
If there is nothing significant to report during this reporting period, state "Nothing to Report."

Describe ways in which the project made an impact, or is likely to make an impact, on commercial technology or public use, including:

- *transfer of results to entities in government or industry;*
- *instances where the research has led to the initiation of a start-up company; or*
- *adoption of new practices.*

Nothing to report

What was the impact on society beyond science and technology?
If there is nothing significant to report during this reporting period, state "Nothing to Report."

Describe how results from the project made an impact, or are likely to make an impact, beyond the bounds of science, engineering, and the academic world on areas such as:

- *improving public knowledge, attitudes, skills, and abilities;*
- *changing behavior, practices, decision making, policies (including regulatory policies), or social actions; or*
- *improving social, economic, civic, or environmental conditions.*

Nothing to report

5. **CHANGES/PROBLEMS:** *The PD/PI is reminded that the recipient organization is required to obtain prior written approval from the awarding agency grants official whenever there are significant changes in the project or its direction. If not previously reported in writing, provide the following additional information or state, "Nothing to Report," if applicable:*

Nothing to report

Actual or anticipated problems or delays and actions or plans to resolve them
Describe problems or delays encountered during the reporting period and actions or plans to resolve them.

The HRPO review process was extended, extending the timeline of the project. We have in the course of less than a year, prescreened individuals from other cohorts, who had provided consent to be re-contacted, for GWI autoantibody panel developed by Dr. Donia. In addition, we have patients from GWI clinic is at NSU that have expressed interest and are being screened for study eligibility at this time. Post HRPO, position was interviewed and hired. We are currently training the individual who is a Masters level trained Nurse practitioner with years of experience in clinical research

Changes that had a significant impact on expenditures

Describe changes during the reporting period that may have had a significant impact on expenditures, for example, delays in hiring staff or favorable developments that enable meeting objectives at less cost than anticipated.

Nothing to report

Significant changes in use or care of human subjects, vertebrate animals, biohazards, and/or select agents

Describe significant deviations, unexpected outcomes, or changes in approved protocols for the use or care of human subjects, vertebrate animals, biohazards, and/or select agents during the reporting period. If required, were these changes approved by the applicable institution committee (or equivalent) and reported to the agency? Also specify the applicable Institutional Review Board/Institutional Animal Care and Use Committee approval dates.

Significant changes in use or care of human subjects

Nothing to report

Significant changes in use of biohazards and/or select agents

Nothing to report

6. PRODUCTS: *List any products resulting from the project during the reporting period. If there is nothing to report under a particular item, state "Nothing to Report."*

- **Publications, conference papers, and presentations**
  Report only the major publication(s) resulting from the work under this award.

  Journal publications. *List peer-reviewed articles or papers appearing in scientific, technical, or professional journals. Identify for each publication: Author(s); title; journal; volume: year; page numbers; status of publication (published; accepted, awaiting publication; submitted, under review; other); acknowledgement of federal support (yes/no).*

Nothing to report

  Books or other non-periodical, one-time publications. *Report any book, monograph, dissertation, abstract, or the like published as or in a separate publication, rather than a periodical or series. Include any significant publication in the proceedings of a one-time conference or in the report of a one-time study, commission, or the like. Identify for each one-time publication: author(s); title; editor; title of collection, if applicable; bibliographic information; year; type of publication (e.g., book, thesis or dissertation); status of publication (published; accepted, awaiting publication; submitted, under review; other); acknowledgement of federal support (yes/no).*

Nothing to report

  Other publications, conference papers and presentations. *Identify any other publications, conference papers and/or presentations not reported above. Specify the status of the publication as noted above. List presentations made during the last year (international, national, local societies, military meetings, etc.). Use an asterisk (*) if presentation produced a manuscript.*

Nothing to report

- **Website(s) or other Internet site(s)**
  List the URL for any Internet site(s) that disseminates the results of the research activities. A short description of each site should be provided. It is not necessary to include the publications already specified above in this section.

Nothing to report

- **Technologies or techniques**
  Identify technologies or techniques that resulted from the research activities. Describe the technologies or techniques were shared.

Nothing to report

- **Inventions, patent applications, and/or licenses**
  Identify inventions, patent applications with date, and/or licenses that have resulted from the research. Submission of this information as part of an interim research performance progress report is not a substitute for any other invention reporting required under the terms and conditions of an award.

Nothing to report

- **Other Products**
  Identify any other reportable outcomes that were developed under this project. Reportable outcomes are defined as a research result that is or relates to a product, scientific advance,

or research tool that makes a meaningful contribution toward the understanding, prevention, diagnosis, prognosis, treatment and /or rehabilitation of a disease, injury or condition, or to improve the quality of life. Examples include:

- *data or databases;*
- *physical collections;*
- *audio or video products;*
- *software;*
- *models;*
- *educational aids or curricula;*
- *instruments or equipment;*
- *research material (e.g., Germplasm; cell lines, DNA probes, animal models);*
- *clinical interventions;*
- *new business creation; and*
- *other.*

Nothing to report

7. PARTICIPANTS & OTHER COLLABORATING ORGANIZATIONS

What individuals have worked on the project?

Provide the following information for: (1) PDs/PIs; and (2) each person who has worked at least one person month per year on the project during the reporting period, regardless of the source of compensation (a person month equals approximately 160 hours of effort). If information is unchanged from a previous submission, provide the name only and indicate "no change".

Example:

Name: *Nancy Klimas*
Project Role: *Principal Investigator*
Researcher Identifier: *eCommons: nklimas*
Nearest person month worked: *1.0*
Contribution to Project: *No Change*

Name: *Mary Ann Fletcher*
Project Role: *Laboratory Associate Director*
Researcher Identifier (e.g. ORCID ID): *eCommons: mFletcher*
Nearest person month worked: *0.60*
Contribution to Project: *Overseeing the establishment of standard operating procedures for the collection, shipping, accession, and*

	specimen processing for both the biorepository and the standard lab panel described in the narrative

Name:	*Maria Abreu*
Project Role:	*Laboratory Associate Director*
Researcher Identifier:	*eCommons: mabreu*
Nearest person month worked:	*0.60*
Contribution to Project:	*Establishment of standard operating procedures for the collection, shipping, accession, and specimen processing for both the biorepository and the standard lab panel described in the narrative*

Name:	*Abou Donia*
Project Role:	*Collaborator/Co-investigator*
Researcher Identifier:	
Nearest person month worked:	*0.60*
Contribution to Project:	*Establishment of standard operating procedures for the collection, shipping, accession, and specimen processing for both the biorepository and the standard lab panel described in the narrative*

Has there been a change in the active other support of the PD/PI(s) or senior/key personnel since the last reporting period?

Please see active other support for Dr. Klimas under appendice.

What other organizations were involved as partners?
Nothing to report.

Organization Name: Duke University
Location of Organization: Durham, NC
Partner's contribution to the project: Collaborator (Laboratory)

8. SPECIAL REPORTING REQUIREMENTS

COLLABORATIVE AWARDS: *For collaborative awards, independent reports are required from BOTH the Initiating Principal Investigator (PI) and the Collaborating/Partnering PI. A duplicative report is acceptable; however, tasks shall be clearly marked with the responsible PI and research site. A report shall be submitted to* ***https://ers.amedd.army.mil*** *for each unique award.*

QUAD CHARTS: *If applicable, the Quad Chart (available on https://www.usamraa.army.mil) should be updated and submitted with attachments.*

9. APPENDICES: *Attach all appendices that contain information that supplements, clarifies or supports the text. Examples include original copies of journal articles, reprints of manuscripts and abstracts, a curriculum vitae, patent applications, study questionnaires, and surveys, etc.*

ACTIVE

W81XWH1820062 (Klimas) 9/30/2018-9/29/2022 2.4 calendar
The Gulf War Illness Clinical Trials and Interventions Consortium (GWICTIC)
Role: PI

GW170055 (Sullivan) 9/01/2018-8/31/2021 0.24 calendar
Boston Biorepository, Recruitment and Integrative Network (BBRAIN)
Role: Co-I

W81XWH1810660 (Nathanson) 9/30/2018-9/29/2021 0.36 calendar
Immunomodulation in GWI
Role: Co-I

W81XWH-17-1-0334 (Klimas) 09/30/2017-09/29/2020 0.6 calendar
The Use of B-Cell Depletion Therapy (BCDT) in Gulf War Illness: A Phase 1/2 Study
Role: PI

W81XWH-16-GWIRP (Fletcher-Klimas) 09/01/2017-04/30/2020 0.36 calendar
VA Foundation
Measurement of Biomarkers in Samples Collected in a Coenzyme Q10 Treatment Trial in Gulf War Illness and Control Subjects.
Role: PI

W81XWH-17-2-0063 (Salgueiro) 09/30/2017-09/29/2020 0.6 calendar

Growth Hormone-Releasing Hormone (GHRH) Antagonist: Evaluation of Beneficial Effects for Gulf War Illness
Role: Co-I

W81XWH-17-1-0640 (Nathanson) 09/15/2017-09/14/2020 0.36 calendar
Genomic approach to find female-specific mechanisms of GWI pathobiology
Role: Co-I

W81XWH-161-1-0632 (Craddock) 09/30/2016-09/29/2020 0.24 calendar
Disentangling the Effects of PTSD from GWI for Improved Diagnostics and Treatments
Role: Co-I

W81XWH-161-1-0552 (Craddock) 09/30/2016-09/29/2020 0.24 calendar
Improving Diagnostics and Treatments for GWI Females by Accounting for the Effects of PTSD
Role: Co-I

W81XWH-16-1-0678 (Grant) 09/30/2016-09/29/2020 0.6 calendar
Persistently elevated somatic mutation as a biomarker for clinically relevant exposures in GWI
Role: Co-I

W81XWH-13-2-0085 (Morris) NCE 09/30/2013 – 03/31/2020 1.2 calendar
Understanding Gulf War Illness: An Integrative Modeling Approach
Role: Co-PI

W81XWH13-2-0072 (Sullivan) 09/30/13-09/29/2020 No measurable effort in NCE
Brain-Immune Interactions as the Basis of Gulf War Illness: Gulf War Illness Consortium (GWIC)
Role: Co-I

R21AI142717 (Abdullah/Crawford/Klimas) 12/21/2018-11/31/2020 0.6 calendar
Application of lipidomics to identify biomarkers of immune and mitochondrial disturbances in Chronic Fatigue Syndrome
Role: Co-PI

W81XWH-19-1-0767 (Sullivan) 09/15/2019 – 09/14/2022 0.6 calendar
Defining and Characterizing GWI Pathobiology using Longitudinal Brain Imaging Biomarkers of White Matter Integrity and Hemodynamic Response
Role: Co-I

W81XWH-15-1-0537 (Klimas) 07/01/2015 – 9/29/20 0.6 calendar
VA Foundation
Testing the Model: A Phase I/II Randomized Double Blind Placebo Control Trial of Therapeutics: Liposomal Glutathione and Curcumin
Role: PI

I01CX001923-01 (Chaterjee) 04/1/2019-04/01/2023 0.6 calendar
VA Merit
CMA: Immune/Inflammatory Priming in Exacerbating Responses to GWVI Stressors: Implications for GWVI Treatments
Role: Co-I

I01CX001480-01 07/01/2016 – 08/30/2020 0.72 calendar
VA Merit GWI $1,029,000
A Randomized, Double-blind Placebo-controlled Phase III Trial of Coenzyme Q10 in Gulf War Illness
Role: PI

I01CX001237-01A1 (Klimas) 08/01/2015–12/01/2020 0.4 calendar
VA Merit
A Translational Medicine Approach to Gulf War Illness: From Cells to Therapy
Role: PI

I01CX001050-01A1 (Klimas) 7/1/2014-5/01/2020 0.6 calendar
VA Merit
Women vs. Men with GWI: Differences in Computational Models and Therapeutic Target
Role: PI