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TITLE: Neurovascular Dysregulation in Myalgic Encephalomyelitis/Chronic Fatigue Syndrome

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14. ABSTRACT

Rationale: Myalgic encephalomyelitis/chronic fatigue syndrome (ME/CFS) is a common and clinically devastating disorder whose pathogenesis is poorly understood. ME/CFS may affect as many as 2.5 million people in the United States. Veterans who were deployed to the Persian Gulf during the Gulf War have a higher prevalence of ME/CFS and the related condition fibromyalgia (FM) compared to non-deployed Veterans. Furthermore, almost a quarter of the Veterans of the Gulf War developed chronic multisystem illness/Gulf War illness (CMI/GWI), which has many overlapping symptoms with ME/CFS. The underestimated direct and indirect cost of ME/CFS to society may approach $23 billion per year.

There is substantial clinical overlap among FM, postural orthostatic tachycardia syndrome (POTS) and ME/CFS. Both FM and POTS have a high prevalence of small fiber neuropathy (SFN) diagnosed by epidermal biopsy, which may be responsible for symptoms. The prevalence of SFN in ME/CFS is not known.

Using a maximum invasive upright exercise test (iCPET), which simultaneously measures ventilation, pulmonary and systemic gas exchange and hemodynamics, we have found that vascular dysregulation and exertional intolerance are highly prevalent in patients with ME/CFS. ***The purpose of this project is to link vascular dysregulation during exercise to small fiber autonomic neuropathy and exercise intolerance in a large cohort of ME/CFS compared to normal controls.***

Hypothesis: Exertional intolerance in ME/CFS is related to neurovascular dysregulation.

Innovative aspects: This will be the first study of ME/CFS to link vascular dysregulation during exercise to SFN. Our innovative approach will attempt to link abnormal neuroanatomy (SFN by epidermal biopsy) to vascular dysregulation (by iCPET) as an underlying mechanism for exercise intolerance for ME/CFS. Furthermore, we aim to deep phenotype ME/CFS, with particular attention to diagnostic vascular and neural subgroups.

Proposed research and study design: We will analyze more than 2000 clinically indicated iCPET's performed at an academic medical center between 2011 and 2020 for unexplained exertional intolerance. Patients who meet the Institute of Medicine (IOM) criteria for ME/CFS, and do not have other comorbidities, will be analyzed in terms of iCPET vascular dysregulation variables and neurite density to obtain the prevalence of vascular dysregulation and SFN in ME/CFS. We will determine if vascular dysregulation (peak right atrial pressure, CO/VO2 slopes, peak Ca-vO2/[Hb] and peak systemic vascular resistance) is predicted by neurite density in ME/CFS vs normals by regression analysis, controlling for age, gender and other variables. Next, a network analysis of all iCPET variables, neurite percentiles and clinical characteristics will be performed. The threshold of the correlation coefficient and the p-value will be determined by bootstrap and multiple test correction. Unsupervised analysis will be performed to explore the potential vascular and neuroanatomical ME/CFS subgroups, according to iCPET hemodynamic and gas exchange variables, neurite density and clinical variables. These include analysis such as various clustering algorithms, principal component analysis (PCA), and t-distributed stochastic neighbor embedding (t-SNE).

Expected results: We predict we will find a high prevalence of vascular (venous and arteriolar) dysregulation and SFN in ME/CFS patients, and the two will be causally linked. We believe that this unique marrying of vascular pathophysiology during exercise to abnormal neuroanatomy will explain exertional intolerance in ME/CFS. We believe that this finding will be relevant to CMI/GWI and fatigue to the civilian and military populations.

Foundation for future projects: These results could set the foundation for a new diagnostic classification of ME/CFS based on vascular and neural anatomic abnormalities. Deep phenotyping and identification of the pathophysiology underlying exercise intolerance in ME/CFS will lead to targeted therapies which will have relevance to CMI/GWI.

15. SUBJECT TERMS
ME/CFS, neurovascular dysregulation, chronic fatigue syndrome, preload failure, small fiber neuropathy

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1. Introduction

Myalgic Encephalomyelitis/Chronic Fatigue Syndrome (ME/CFS) is a devastating and poorly understood condition that affects up to 2.5 million people in the United States and more than 5,000 Veterans. ME/CFS is associated with military deployment and is a disease with substantial clinical overlap with other conditions of interest to CDMRP, such as chronic multisymptom illness/Gulf War illness (CMI/ GWI). Despite the impact of this disease, little is known about the pathophysiology, diagnosis and treatment. There is a large subgroup of ME/CFS patients who also have small fiber neuropathy (SFN). SFN is indicated by a reduced number of nerves shown during a skin biopsy. The prevalence and relevance of blood vessel dysfunction to SFN and exercise intolerance, as well as its relationship with the blood flow and gas exchange throughout the lungs and the rest of the body in ME/CFS, are unknown. Using a maximum invasive upright exercise test (iCPET) and morphological and/or functional testing for SFN, this project's main objective is to link abnormal neuroanatomy to vascular dysregulation as an underlying mechanism for exercise intolerance in ME/CFS.

2. Keywords

ME/CFS, neurovascular dysregulation, chronic fatigue syndrome, preload failure, small fiber neuropathy

3. Accomplishments

Specific Aim 1: Determine the prevalence of myalgic encephalomyelitis/ chronic fatigue syndrome (ME/CFS) in patients who undergo an invasive cardiopulmonary exercise test (iCPET) for the evaluation of exertional intolerance.	**Timeline**	**Status**
Major Task 1: Obtain IRB and HRPO approvals	**Months**	COMPLETE
Subtask 1: Elaboration and submission of IRB initial review documents.	-6 to-4	COMPLETE
Subtask 2: Elaboration and submission of HRPO documents.	-4 to 0	COMPLETE
Milestone(s) Achieved: IRB and HRPO approvals.	0	COMPLETE
Major Task 2: Identify patients with ME/CFS from the computer records of the patients who have undergone an iCPET for the evaluation of unexplained exertional intolerance.	**Months**	COMPLETE
Subtask 1: Obtain the clinical computer records of all the patients who have undergone an iCPET for unexplained exertional intolerance evaluation from 2011 to 2020 at Brigham and Women's Hospital.	0 to 1	COMPLETE
Subtask 2: Perform a RPDR search of electronic medical records for fatigue related terms of the identified patients.	0 to 1	COMPLETE
Subtask 3: Of the identified patients, determine who meet the Institute of Medicine (IOM) criteria for ME/CFS.	1 to 2	COMPLETE

Milestone(s) Achieved: Determination of the prevalence of ME/CFS in patients who underwent a clinically indicated iCPET for the evaluation of exertional intolerance between 2011 and 2020.	2	COMPLETE
Major Task 3: Of patients who have been identified after Major Task 1, exclude active clinically determined comorbidities from the electronic medical records.		COMPLETE
Subtask 1: Exclude patients with precapillary and postcapillary pulmonary hypertension (PH) and all forms of heart failure.	2 to 3	COMPLETE
Subtask 2: Exclude patients with active and treated cancer, morbid obesity (BMI < 40 kg/m2), non-controlled asthma, and severe anemia (Hb < 9 g/dL).	2 to 3	COMPLETE
Subtask 3: Exclude patients with any other unexpected comorbidity found during chart review that might be a potential cofounder, as determined by the principal investigator (PI).	2 to 3	COMPLETE
Milestone(s) Achieved: Identification of a cohort of ME/CFS free of clinically apparent comorbidities.	3	COMPLETE
Major Task 4: Of the patients who remain after Major Task 2, exclude patients with comorbidities determined by the iCPET, elite athletes and incomplete exercise data.		COMPLETE
Subtask 1: Exclude patients with submaximum testing: maximum heart rate achieved < 85 % predicted for age or a peak respiratory exchange ratio (RER) < 1.05.	3 to 3.5	COMPLETE
Subtask 2: Exclude patients with primary pulmonary mechanical limitation to exercise: minute ventilation/maximum voluntary ventilation > 0.7 at the anaerobic threshold.	3 to 3.5	COMPLETE
Subtask 3: Exclude patients with precapillary and postcapillary PH.	3 to 3.5	COMPLETE
Subtask 4: Exclude patients with heart failure with preserved ejection fraction.	3 to 3.5	COMPLETE
Subtask 5: Exclude patients with exercise precapillary and postcapillary PH.	3 to 3.5	COMPLETE
Subtask 6: Exclude elite athletes with VO2 max > 120%.	3 to 3.5	COMPLETE
Subtask 7: Exclude patients with incomplete data.	3 to 3.5	COMPLETE
Milestone(s) Achieved: Identification of a cohort of ME/CFS free of iCPET determined comorbidities.	3 to 3.5	COMPLETE
Major Task 10: First manuscript write up and publication of results		INCOMPLETE

Subtask 1: Manuscript about ME/CFS prevalence in patients who undergo iCPETs, selection of the journal and publication.	3.5-4.5	INCOMPLETE
Milestone(s) Achieved: First manuscript published in high impact journal.	6.5	INCOMPLETE
Major Task 5: Identify normal controls.		COMPLETE
Subtask 1: Identify normal subjects with VO2, CO, and systemic oxygen extraction (SOE) (Ca-vO2/[Hb]) > 80% of predicted.	4.5-5	COMPLETE
Subtask 2: Exclude subjects with comorbidities as performed in Major Tasks 2 and 3.	5-6	COMPLETE
Milestone(s) Achieved: Identification of a comparator group of normal controls.	6	COMPLETE
Specific Aim 2: Document the prevalence of vascular dysregulation during exercise in ME/CFS.		COMPLETE
Major Task 6: Identify the prevalence of venous dysregulation during exercise in ME/CFS patients.		COMPLETE
Subtask 1: Identify patients with preload failure, defined as a peak right atrial pressure (RAP) < 6.5 mmHg.	6 to 6.5	COMPLETE
Subtask 2: Compare iCPET data of these patients to normal controls.	6 to 6.5	COMPLETE
Milestone(s) Achieved: Determination of the prevalence of venous dysregulation.	6.5	COMPLETE
Major Task 7: Identify ME/CFS patients with evidence of arteriolar dysregulation during exercise.		COMPLETE
Subtask 1: Identify arteriolar dysregulation by assessing the cardiac output/oxygen uptake slopes (CO/VO2 slopes), and subdividing them: low, normal and high; and identifying SOE (Ca-vO2max/[Hb]) and systemic vascular resistance (SVR).	6.5 to 7	COMPLETE
Subtask 1: Identify subsets of ME/CFS patients with peripheral left-to-right shunting defined as high CO/VO2 slope, poor SOE (Ca-vO2max/[Hb] < 0.8) and low SVR (SVR < 800 dynes).	6.5 to 7	COMPLETE
Subtask 2: Compare iCPET data of these patients to normal controls.	7 to 8	COMPLETE
Milestone(s) Achieved: Identification of arteriolar dysregulation prevalence in ME/CFS patients.	8	COMPLETE
Major Task 10: Second manuscript write up and publication of results		INCOMPLETE
Subtask 1: Manuscript about vascular dysregulation during exercise in ME/CFS elaboration, selection of the journal, and publication.	8 to 10	INCOMPLETE

Milestone(s) Achieved: Second manuscript published in high impact journal.	12	INCOMPLETE
Specific Aim 3: Document the prevalence of small fiber neuropathy (SFN) in ME/CFS.		INCOMPLETE
Major Task 8: Identify SFN prevalence in ME/CFS patients.		COMPLETE
Subtask 1: Of the patients identified in Major Task 3, identify those with an epidermal skin biopsy, specially stained by PGP9.5-immunolabel for neurite density and corresponding neurite percentile, and identify patients with definite, probable and without SFN.	10 to 12	COMPLETE
Milestone(s) Achieved: Identification of SFN prevalence and neuroanatomic phenotypes in ME/CFS patients.	12	COMPLETE
Major Task 10: Third manuscript write up and publication of results		INCOMPLETE
Subtask 1: Manuscript about SFN prevalence and neuroanatomical phenotypes in ME/CFS elaboration, selection of the journal and publication.	12 to 14	INCOMPLETE
Milestone(s) Achieved: Third manuscript published in high impact journal.	16	INCOMPLETE
Specific Aim 4: Determine if neurovascular dysregulation underlies exercise intolerance in ME/CFS.		
Major Task 9: Statistically relate SFN to vascular dysregulation during exercise in ME/CFS.		INCOMPLETE
Subtask 1: Determine if vascular dysregulation (peak RAP, CO/VO2 slopes, peak Ca-vO2/[Hb] and peak SVR) is predicted by neurite density in ME/CFS vs normals by regression analysis, controlling for age, gender and other variables.	14 to 16	INCOMPLETE
Subtask 2: Perform network correlation analysis to link vascular dysregulation by iCPET to neurite percentiles and clinical characterizes,	16 to 18	INCOMPLETE
Subtask 3: Perform an unsupervised analysis to explore the potential ME/CFS vascular and neuroanatomical ME/CFS subgroups by various clustering algorithms, principal component analysis (PCA), and t-distributed stochastic neighbor embedding (t-SNE).	18 to 20	INCOMPLETE
Milestone(s) Achieved: Demonstrate that neurovascular dysregulation underlies exercise intolerance in ME/CFS.	20	INCOMPLETE
Major Task 10: Final manuscript write up and publication of results		INCOMPLETE
Subtask 1: Manuscript about Neurovascular dysregulation in ME/CFS elaboration, selection of the journal and publication.	20-24	INCOMPLETE

Milestone(s) Achieved: Final manuscript published in high impact journal.	24	INCOMPLETE

- **What opportunities for training and professional development has the project provided?** – Nothing to report
- **How were the results disseminated to communities of interest? -** Results have yet to be disseminated to communities of interest.
- **What do you plan to do during the next reporting period to accomplish the goals? -**

  We have completed chart review, data entry, and review of the database in accordance with the major task criteria listed above. During the next period, we will perform biostatistical analysis (analysis of all iCPET variables, neurite percentiles and clinical characteristics, bootstrap and multiple test correction to determine threshold of correlation coefficient and the p value, and unsupervised analysis for exploration of potential ME/CFS vascular and neuroanatomical ME/CFS subgroups). We will then write up and publish these results.

4. Impact

Nothing to report

5. Changes/Problems

Nothing to report

6. Products

Nothing to report

7. Participants & Other Collaborating Organizations

Name	Dr. David Systrom
Project Role	PD/PI
Researcher Identifier (e.g. ORCID ID):	0000-0002-9610-6330
Nearest person month worked	6 months
Contribution to Project	Dr. Systrom began as an "other significant contributor" but has since taken over as PD/PI of the study. The ME/CFS database is largely composed of his patients and he is viewed as a leader in this field of study.
Name	Dr. Rosa Pari Nana
Project Role	Previous PD/PI
Nearest person month worked	6 months
Contribution to Project	Dr. Pari Nana proposed and initially crafted this study. She oversaw all applications and IRB approval.
Name	Dr. Ronald Tompkins
Project Role	Other significant contributor
Nearest person month worked	6 months

Contribution to Project	Dr. Tompkins has provided immeasurable advice, guidance, and support (moral and intellectual) to the study as he has high hopes for the impact of this project.
Name	Mary Catherine Stovall
Project Role	Research Coordinator
Nearest person month worked	6 months
Contribution to Project	Ms. Stovall has performed work involved with patient chart review and data entry in the ME/CFS database. She also took lead on IRB correspondence for the study.
Name	Johanna Squires
Project Role	Research Coordinator
Nearest person month worked	2 months
Contribution to Project	Ms. Stovall has performed work involved with patient chart review and data entry in the ME/CFS database. She also performed preliminary statistical analysis on the findings.
Name	Sarah Miller
Project Role	Research Coordinator
Nearest person month worked	1 month
Contribution to Project	Ms. Miller performed work involving patient chart review and data entry in the ME/CFS database.
Name	Arabella Warren
Project Role	Research Coordinator
Nearest person month worked	3 months
Contribution to Project	Ms. Warren performed work involving patient chart review and data entry in the ME/CFS database.
Name	Alex Kingston
Project Role	Research Coordinator
Nearest person month worked	3 months
Contribution to Project	Mr. Kingston performed work involving patient chart review and data entry in the ME/CFS database.

- **Has there been a change in the active other support of the PD/PI(s) or senior/key personnel since the last reporting period? –** The PD/PI of this study is no longer Dr. Pari Nana and is now Dr. David Systrom.
- **What other organizations were involved as partners?** - Nothing to report

8. Special Reporting Requirements

Not applicable