



MRC-funded research projects

The MRC is currently funding the following projects relating to CFS/ME:

MR/J002895/1

PI: Professor Anne McArdle, University of Liverpool

Title: Determination of mitochondrial function and cytokine production in skeletal muscle of patients with CFS.

Start Date: 01/05/2012

End Date: 30/04/2015

Award Amount: £252,030.40

Lay Summary

Chronic fatigue syndrome (CFS) is a severely debilitating illness of uncertain cause. CFS is characterised by prolonged, debilitating fatigue that can be triggered by minimal activity (NICE, 2010). The fatigue is accompanied symptoms which can include painful muscles and joints, disordered sleep, gastric disturbances and cognitive impairment and is sometimes associated with depression. CFS can affect people of any age but is most common between the ages of 25 and 45. Evidence suggests between 150,000 and 250,000 people are affected in the UK. The effect of CFS on quality of life is substantial, with some individuals becoming housebound, employment becoming difficult or impossible, disrupted education in younger sufferers and thus represents a substantial effect on the quality of life for people with the condition, their families and carers.

The mechanisms by which an initial event leads to the chronic debilitating muscle fatigue and pain are unknown. The time required for diagnosis (typically 4 - 6 months) further complicates the identification of the factor(s) responsible for initiation of the illness. Irrespective of the factor(s) which initiates the illness, reversal of the severely debilitating fatigue which ensues remains the most promising form of treatment.

A number of studies have suggested that there is a defect in the energy producing components of muscle cells, known as mitochondria but, although this is core to understanding muscle fatigue in patients with CFS, the presence of abnormal mitochondria in muscle of patients with CFS remains the subject of considerable debate as other studies have failed to demonstrate a defect. The reasons for such different findings are likely due to the previously limited methods of analysis for mitochondrial function with a lack of availability of appropriate and sensitive techniques to determine mitochondrial function directly in human muscle fibres. However, a new technique to study mitochondria in muscle fibres in situ from humans has now been developed and is established in our laboratory.

We hypothesise that the application of these newly developed techniques will demonstrate that skeletal muscle mitochondria in patients with CFS are dysfunctional and that this results in muscle fatigue. The dysfunctional

mitochondria then activate a process which leads to a chronic, low grade inflammation, commonly reported in patients with CFS, which in turn results in further mitochondrial abnormalities and the establishment of a vicious circle of events. Understanding the processes by which muscle fatigue occurs will lead to optimal interventions that break this vicious circle and improve muscle function and wellbeing of individuals.

Technical Summary

Chronic Fatigue Syndrome (CFS) is a severely debilitating illness of uncertain cause, characterised by prolonged, debilitating fatigue. Reversal of the severely debilitating fatigue which ensues remains the most promising treatment. The presence of abnormal mitochondria in muscle of patients with CFS remains the subject of considerable debate. The reasons for such different findings are likely due to the previous lack of availability of appropriate and sensitive techniques to determine mitochondrial function directly in muscle fibres. However, to study mitochondrial function in muscle fibres in situ from humans, a method of isolating bundles of muscle fibres and permeabilisation with saponin has been developed. This technique is established in our laboratories and will lead to a definitive answer regarding a mitochondrial defect in muscles of patients with CFS. Chronic ROS generation by muscle mitochondria is proposed to result in chronic activation of NFkB and subsequent lowgrade inflammation. We further hypothesise that activation of NFkB results in muscle becoming a major source of systemic pro-inflammatory cytokines, resulting in further mitochondrial abnormalities and the establishment of a vicious circle of events. Interventions that modify mitochondrial ROS generation or NFkB activation in muscles will reduce systemic inflammation, break this vicious circle and improve muscle function. This application is a new collaboration between basic scientists at the University of Liverpool, experts on diet and cognitive function at the University of Leeds and a consultant in Infectious disease and Tropical medicine with a special interest in CFS/ME. We will apply a novel technique to examine mitochondrial function in muscle cells in situ and determine the role of muscle in the production of inflammatory mediators.

MR/J002712/1**PI:** Professor Julia Newton, Newcastle University**Title:** Understanding the pathogenesis of autonomic dysfunction in chronic fatigue syndrome and its relationship with cognitive impairment**Start Date:** 01/06/2012**End Date:** 31/05/2015**Award Amount:** £454,573.48**Lay Summary**

Chronic fatigue syndrome (CFS) occurs in 0.2-0.4% of Europe's population, can affect all ages and currently its cause is unclear. Abnormality of the autonomic nervous system is recognised in over three quarters of those with CFS and is a plausible physiological mediator of the symptoms that are characteristic of CFS and fatigue in other chronic diseases. Autonomic nervous system dysfunction is characterised by symptoms of dizziness and lightheadedness when standing up, symptoms that we have shown to be present in nearly 90% of people with CFS, and the severity of which have been shown to predict the ability of CFS patients to function (more so than the severity of fatigue). Despite this, the mechanisms by which autonomic dysfunction arises in those with chronic fatigue syndrome are not understood and as a result treatments limited.

This study fills this gap by setting out to explore what leads to autonomic dysfunction in CFS using novel methodologies particularly whether it is upstream (related to abnormalities in centres in the brain that control the autonomic nervous system) or downstream (due to a peripheral volume or vascular problem) in origin. In non-CFS diseases autonomic dysfunction has also been shown to be associated with cognitive impairment. Over 80% of those with CFS describe problems with memory and concentration, so this study will also determine the relationship between autonomic dysfunction and these cognitive problems frequently found in those with CFS. Utilising the enormous resource created by this integrated study, the programme will look to develop diagnostic biomarkers.

The programme has two complementary phases: 1) an exploratory study that utilises ground breaking dynamic MR modalities that will allow study of brain function in CFS and how this relates to autonomic and cognitive function. 2) a downstream study which combines a number of work packages to define the relative contribution of cardiac and vascular function in autonomic dysfunction.

Understanding the mechanisms that lead to autonomic dysfunction in those with CFS will be a paradigm shift. This programme will lay a foundation for research by the applicant and others that will enable a future set of diagnostic tools, system based explanations of dysfunction, a new generation of therapies and ultimately clinical protocols that will counter the biological processes that underpin fatigue in a range of diseases.

This proposal will use state of the art techniques such as dynamic brain FMRI to measure cerebral blood flow during the autonomic nervous system stressor of the valsalva manoeuvre (considered to be a test of cerebral autoregulation) to understand the mechanisms that lead to autonomic dysfunction and the associated cognitive impairment seen in the majority of those with CFS.

This project will directly benefit patients through improving our understanding of how autonomic dysfunction arises in CFS and how it associates with cognitive function. This enhanced understanding will lead to the development of targeted appropriate treatments for clinical trials which will be aimed at reversing these abnormalities.

Technical Summary

Chronic Fatigue Syndrome (CFS) is a debilitating disease that can affect all ages and profoundly influences a sufferer's ability to function. Despite its impact, the cause of CFS remains unknown and there are no effective treatments. One consistent theme in the CFS literature is of compromise of the autonomic nervous system which has led to the concept that abnormality of regulation of the autonomic nervous system (autonomic dysfunction (AD)) underpins the pathogenesis and/or clinical expression of CFS. AD has been associated with cognitive impairment and risk of cognitive decline in non-CFS groups. Defining the pathogenesis of AD and its relationship with cognitive impairment would be of immense value for both the study of the pathogenesis and treatment of CFS, and the clinical management of fatigued patients.

Hypothesis Autonomic dysfunction in CFS arises due to combination of central ('upstream') and peripheral ('downstream') abnormalities in blood pressure regulation. This leads to symptoms of brain hypoperfusion which is a cause of cognitive dysfunction. The aim of this study is to define the underlying physiological abnormalities that lead to autonomic dysfunction in CFS and its relationship with cognitive impairment.

Methodology Pathogenesis of AD will be determined using novel state of the art MR technologies developed for this application in CFS patients with an autonomic phenotype (with newly diagnosed and established disease) compared to CFS without autonomic phenotype and sedentary controls

Deliverables Having a comprehensive understanding of the pathogenesis of AD in CFS and its relationship with cognitive impairment will lead to targeted clinical trials that will improve autonomic symptoms, enhance functional ability and reduce cognitive impairment.

MR/J002720/1**PI:** Dr Wan Ng, Newcastle University**Title:** Identifying the biological fingerprints of fatigue**Start Date:** 01/01/2012**End Date:** 31/12/2014**Award Amount:** £451,572.73**Lay Summary**

Context of the research:

Chronic fatigue syndrome (CFS) affects 1 in 300 people in the UK and is associated with a significant healthcare-related cost to the society. Severe, persistent fatigue is a key symptom of CFS and a major factor leading to loss of productivity in this illness. The cause of CFS and the underlying biological mechanisms of fatigue are poorly understood. As a result, accurate diagnosis of CFS can be difficult and effective treatment is not available. A growing body of evidence suggests that CFS may be linked to a faulty immune system. However, in what ways the immune system is not working properly is not clear. Therefore, by uncovering the abnormalities of the immune system in CFS in detail, it will help make the diagnosis of CFS easier and more accurate as well as give us clues to develop effective treatment of this condition.

Since the diagnosis of CFS is unreliable at present, a group of "CFS" patients may in fact consist of individuals with different diseases. This presents a major obstacle to the progress of CFS research and may also explain why data from different CFS research studies are often conflicting. Therefore, in order to better understand the underlying biological mechanisms of CFS, a different approach is needed. Recent data show that intense fatigue is also a common symptom for many chronic conditions. Interestingly, most of these conditions are due to a faulty immune system. Furthermore, research also suggests that severe fatigue in these various conditions is driven by similar biological mechanisms. If so, we may be able to find out the underlying defects of the immune system causing disabling fatigue in CFS by using one of these chronic conditions as a disease model. This is precisely what we will do in this study.

We will carry out a comprehensive analysis of the immune system of a large number of patients with a condition called primary Sjögren syndrome (PSS). We will analyze the data obtained from these experiments to find out what abnormalities of the immune system are linked to fatigue. Since these experiments typically produce a vast amount of data, we will apply statistical methods and mathematical modelling specifically designed to analyse large volumes of biological data (known as bioinformatics and biostatistics) in order to identify the biological "fingerprints" of fatigue. We will then test whether these biological fingerprints of fatigue are present in CFS patients and whether it will help us to diagnose CFS more accurately.

We chose PSS as a disease model for several reasons. (i) PSS and CFS have many shared clinical and biological features including profound fatigue. (ii) Clinical samples from over 550 PSS patients across the UK, as well as their clinical data, are available for this study. Access to such samples is a distinct advantage because this is one of the largest clinical sample collections in PSS in the world. It would be time-consuming, labour-intensive and expensive if we had needed to collect the same amount of clinical samples and accompanied clinical data from fresh. (iii) There are well-established diagnostic criteria for PSS and so this avoids the problem of studying patients with potentially mixed diagnoses as in the case of studying CFS patients.

Aims of the study, potential applications and benefits:

The main objective of this study is to find the biological fingerprints of fatigue. By doing so, it will improve our understanding of the biological mechanisms of fatigue. The data will enable us to develop treatments for the fatigue that plagues so many patients with CFS and other chronic conditions. It will also help us to design a clinical test for the diagnosis of CFS.

Technical Summary

This study aims to identify the biological fingerprints of fatigue using primary Sjogren's syndrome (PSS), an autoimmune condition with several clinical and biological features similar to chronic fatigue syndrome (CFS) including intense fatigue, as a disease model. We will then test whether these biological fingerprints are also present in patients with CFS.

We will perform whole blood gene expression profiling (using genome-wide microarray) and measure serum markers of immune dysregulation (using fluorescent bead-based multiplex technology) of an existing biobanked samples from a large cohort of clinically well-characterized PSS patients (the UK PSS registry). Further information concerning the cohort can be found on the cohort website www.sjogrensregistry.org and Annex 1. The data from these experiments will be analyzed using various bioinformatics techniques in order to identify a biological profile of fatigue, which will be validated using a second blinded test cohort of PSS patients. We will also investigate whether the biological profile changes over time and responds to biological treatment of fatigue in PSS patients. The data from these investigations will then be integrated to identify a set of biomarkers that will maximally discriminate fatigued subjects from non-fatigued individuals. Finally, we will test whether these biomarkers of fatigue are present in CFS patients and if so, whether they can be used to correctly classify CFS from active or sedentary healthy individuals.

MR/J002852/1**PI:** Professor David Nutt, Imperial College London**Title:** Can enhancing SWS improve daytime function in patients with CFS?**Start Date:** 01/04/2012**End Date:** 31/03/2013**Award Amount:** £119,999.60**Lay Summary**

Sleep disturbance is a core symptom of chronic fatigue syndrome (CFS) and has a huge negative impact on daytime function and quality of life. Studies of sleep in the past 10 years have provided evidence that brain mechanisms of sleep regulation, and in particular homeostasis, are disrupted in CFS. Impaired homeostatic mechanisms of sleep result in poor sleep at night and sleepiness and fatigue during the day, contributing to the subjective and objective cognitive impairment seen in these patients. This study will bring together experts in CFS, sleep and psychopharmacology, to study the nature of homeostatic impairment in CFS and its impact on daytime function. We propose to use a pharmacological agent which increases deep restorative sleep (slow wave sleep) which is a marker for homeostatic drive to sleep at night. We will perform a single-dose challenge test in patient with CFS, to ascertain the extent to which this brief and safe pharmacological enhancement of slow wave sleep (and thus of homeostatic mechanisms) will have a significant beneficial impact on daytime impairment. We will include measures of sleepiness, vigilance, memory and subjective well-being. If our results are positive, this will clearly have several potential benefits to CFS sufferers. First, it will underscore the extent to which a major biological function, namely the homeostatic component of the sleep-wake cycle, is impaired in CFS. Second, it will enable us to focus on a specific important brain pathway. Third, it will allow us to evaluate the extent to which patients' daily functions and quality of life are likely to improve following a good night's refreshing sleep. Fourth, our results would direct future major programmes of research into understanding better the underlying sleep disorder in CFS. Finally, the proposed work may suggest potential therapeutic interventions.

Technical Summary

Alterations in slow wave sleep (SWS) and slow wave activity (SWA), the most reliable markers of sleep homeostasis, suggest there may be homeostatic dysregulation in CFS. SWS enhancement improves daytime sleepiness and performance on a number of tasks and the detrimental effects of sleep deprivation on performance can be rescued by administering SWS enhancing drugs. We hypothesised that pharmacological enhancement of SWS may lead to improvements in sleep maintenance and daytime function in CFS patients suffering from non-restorative sleep. This may represent a new avenue for future treatment.

The objective of the research is to compare aspects of daytime performance, notably sleepiness, memory, subjective well-being and fatigue after a night's sleep in which SWS has been enhanced with sodium oxybate in comparison with placebo. This is a randomised, double-blind, placebo-controlled crossover study in patients with CFS. 24 patients will spend two 20-hr periods in the research centre, separated by at least a week, where they will have their sleep recorded overnight. They will be given oral liquid sodium oxybate (3g) or matching placebo in divided doses; 15 minutes prior to usual bedtime and again after 3 hours. Sleep will be recorded continuously until subjects' usual rise time or after a maximum of 10 hours. The following day, assessments of sleep propensity (MSLT) will be made, by the standard method of creating sleep opportunities every 2 hours and measuring time to fall asleep. Tests of vigilance, memory,

visual processing, executive function and subjective experience will be made at intervals during the day. Sleep will be scored using standard methods and spectral analysis will be used to obtain measures of microarchitecture. Subject's daily routines will be measured with actigraphy for the entire study duration.

The results will be published in peer-reviewed journals and more widely in the non-academic community, and will be used to plan future research.

MR/J002739/1**PI:** Dr Carmine Pariante, King's College London**Title:** Persistent Fatigue Induced by Interferon-alpha: A New Immunological Model for Chronic Fatigue Syndrome**Start Date:** 01/03/2012**End Date:** 28/02/2015**Award Amount:** £373,075.15**Lay Summary**

Chronic fatigue syndrome (CFS) is a medical condition in which patients feel persistently and overwhelmingly tired and run down, both physically and mentally. In addition, they have difficulty with concentration, flu-like symptoms and aches and pains. This condition interferes with daily life activity, and, in some patients, is profoundly disabling. Although many years of research have been conducted on CFS, we still do not know what is causing it.

One biological system that is involved in CFS is the "immune system", that is, the system dedicated to fight infections in our body. Indeed, in many cases CFS is triggered by an infection, but then the symptoms continue even after the infection has been eliminated. Specifically, infections are always accompanied by acute fatigue and flu-like symptoms, as a consequence of the infection-driven immune activation; however, in patients with CFS the immune activation and the associated fatigue and flu-like symptoms persist for months or years. Moreover, there is evidence that the immune system is in a state of "hyper-activity" in patients with CFS, as if they were fighting an infective agent, even though they do not have an ongoing infection.

This project aims to understand exactly this process: how the infection and the acute immune activation evolve into CFS, and what are the risk factors that make this process occur in some individuals but not others. Clearly, trying to study this process in subjects experiencing naturally-acquired infections is very difficult, for the unpredictability of these events. In contrast, we want to model the development of CFS by studying a group of patients that have a pre-existing infection (chronic viral hepatitis C, HCV) and that receive a course of treatment (lasting months) with the immune activator, interferon-alpha (IFN-alpha). IFN-alpha is the treatment of choice for HCV infection. Because it activates the immune system, IFN-alpha also induces fatigue and flu-like symptoms in all patients. Moreover, and of particular relevance for this study, a considerable proportion of patients continue to experience debilitating persistent fatigue, and other symptoms that are similar to CFS, for 6 months or even one year after the cessation of IFN-alpha. This phenomenon strikingly resembles CFS, which, as mentioned above, also persists after the infective/immune trigger has been eliminated. Therefore, we are proposing to use IFN-alpha as a model to understand how an immune trigger induces persistent fatigue even when the initial immune trigger is no longer present.

To do this, we will assess these patients throughout the many months of IFN-alpha treatment and at 6 months after cessation of treatment, in order to identify those with persistent "post-IFN-alpha-treatment" fatigue, and understand what biological and clinical changes lead to this outcome. Moreover, we will compare these patients with a group of patients with CFS and with a group of healthy individuals, conducting the same biological and clinical assessment. We will measure changes occurring in blood hormones that are relevant to the immune function, such as "cytokines" and "cortisol". In addition, we will assess changes in measures of well-being, including physical fitness, concentration, sleep and mood.

We are confident that creating and validating this model of CFS will generate a host of future studies aimed at improving the health of people with CFS. For example, we will be able to build a check-list of blood measures that could predict who will, and who will not, develop CFS; we will test novel treatments for "post-IFN-alpha-treatment" fatigue, facilitated by the fact that these patients are homogeneous in their clinical background, and then extend these treatments to patients with CFS; and, finally, we will truly understand what happens in the body during the development of CFS, and thus identify novel therapeutic approaches to interrupt this development.

Technical Summary

We propose to model chronic fatigue syndrome (CFS) by studying patients taking interferon-alpha (IFN-alpha) for chronic viral hepatitis C (HCV) infection. IFN-alpha treatment leads to acute fatigue in the majority of patients. Most importantly, a proportion of patients continue to experience persistent fatigue, together with other CFS-like-symptoms, for many months after the cessation of treatment, that is, in the absence of the pro-inflammatory stimulus. This phenomenon strikingly resembles CFS, which also persists after the viral/immune trigger has been eliminated.

In order to develop this as a model of CFS, in our three-year project we want to:

- 1) assess a cohort (n=100) of patients throughout the IFN-alpha treatment and at 6 months after cessation of treatment, and identify the group who develop the persistent post-treatment fatigue (expected n=50);
- 2) validate this model, by comparing the clinical and biomarkers profiles in patients who experience persistent post-treatment fatigue, patients with CFS (n=50), and healthy controls (n=50);
- 3) identify the risk factors and the biomarkers trajectories (before and during IFN-alpha treatment) that identify those patients who will later experience persistent post-treatment fatigue.

We will measure: fatigue, mood, and other CFS-like symptoms; medical and psychiatric history; childhood and recent stressors; social support; illness and treatment perceptions; physical fitness; quality of life; and occupational function. Moreover, we will measure blood biomarkers: serum cytokines; cortisol at awakening and during the day; and leukocytes gene expression.

The project will build onto an existing pilot study in HCV patients, an established collaboration with Liver Units across London, and the research-led clinical service for CFS patients at King's College Hospital. Thus, the project has great chances of success.