



Understanding, diagnosing, and treating Myalgic encephalomyelitis/chronic fatigue syndrome – State of the art: Report of the 2nd international meeting at the Charité Fatigue Center

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ABSTRACT

Myalgic Encephalomyelitis/Chronic Fatigue Syndrome (ME/CFS) is a devastating disease affecting millions of people worldwide. Due to the 2019 pandemic of coronavirus disease (COVID-19), we are facing a significant increase of ME/CFS prevalence. On May 11th to 12th, 2023, the second international ME/CFS conference of the Charité Fatigue Center was held in Berlin, Germany, focusing on pathomechanisms, diagnosis, and treatment. During the two-day conference, more than 100 researchers from various research fields met on-site and over 700 attendees participated online to discuss the state of the art and novel findings in this field. Key topics from the conference included: the role of the immune system, dysfunction of endothelial and autonomic nervous system, and viral reactivation. Furthermore, there were presentations on innovative diagnostic measures and assessments for this complex disease, cutting-edge treatment approaches, and clinical studies. Despite the increased public attention due to the COVID-19 pandemic, the subsequent rise of Long COVID-19 cases, and the rise of funding opportunities to unravel the pathomechanisms underlying ME/CFS, this severe disease remains highly under-researched. Future adequately funded research efforts are needed to further explore the disease etiology and to identify diagnostic markers and targeted therapies.

1. Introduction

ME/CFS is a frequent and complex disease triggered by various, mostly viral infections [1]. Despite being a severe and common illness, ME/CFS is part of a spectrum of post-infectious diseases that has received little attention from medicine and research for decades. The pandemic has led with Long COVID-19 to a dramatic increase in post-infectious syndromes. It was shown that severe acute respiratory syndrome coronavirus type 2 (SARS-CoV-2) can trigger ME/CFS in a subset of patients [2,3]. Before the emergence of SARS-CoV-2, already an estimated three million people in Europe suffered from ME/CFS, and as a result of the COVID-19 pandemic, many millions more were hit by this devastating disease [4–6]. The most relevant symptoms include mental and physical fatigue, exertional intolerance with post-exertional malaise (PEM), cognitive impairment, orthostatic intolerance, and pain, which often severely interfere with daily life. As of now, the knowledge of pathomechanisms underlying ME/CFS is fragmented, no diagnostic markers have been established, and there is still no approved treatment. The role of immune, vascular, metabolic, and/or autonomic nervous system dysfunction in disease pathogenesis is under intensive investigation [7]. Furthermore, a significant overlap with mechanisms identified in post-COVID syndrome (PCS) has been demonstrated [8–10]. The research initiated worldwide on PCS is bearing great hope for elucidation of ME/CFS pathomechanisms and the development of diagnostic

markers and targeted therapies.

2. ME/CFS belongs to the spectrum of post-COVID syndrome

Yehuda Shoenfeld opened the meeting by giving an expert overview on autoimmunity with regard to the autonomic nervous system focusing on the role of G-protein coupled receptors (GPCR) autoantibodies (AABs), a topic that has been intensively researched recently [11]. Several studies provide evidence that anti-GPCR AABs targeting adrenergic and acetylcholine receptors play a role in ME/CFS, PCS, and postural tachycardia syndrome (POTS) in which autonomic dysregulation is a prominent feature [12,13].

Carmen Scheibenbogen presented the results of an ongoing observational study on the clinical presentation and disease course of ME/CFS triggered by SARS-CoV-2. Of PCS patients presenting with moderate to severe fatigue and exertion intolerance approximately one half fulfilled the Canadian Consensus Criteria (CCC) [14] for the diagnosis of ME/CFS and most patients remained severely affected beyond 18 months of disease onset [2,3]. Furthermore, studies on the association of anti-GPCR AABs with the severity of symptoms, endothelial dysfunction, and candidate biomarkers [9,15] were shown, and an outlook on clinical trials targeting these mechanisms was given.

ME/CFS and PCS share a range of underlying potential abnormalities as outlined in an extensive review by **Anthony L. Komaroff**, including abnormalities pertaining to the central and autonomic nervous system, the immune system, energy metabolism, oxidative stress, cardiopulmonary exercise capacity and the cardiovascular system [16]. Of the identified disease mechanisms, many interact with one another, such as potential viral replication and reactivation, which may lead to oxidative

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and nitrosative stress, in turn leading to cellular senescence. Oxidative stress can also induce mitochondrial dysfunction, which in interaction with viral replication and reactivation results in nitric oxide dioxygenase (NOD)-, leucine-rich repeat (LRR)- and pyrin domain-containing protein 3 (NLRP3)- inflammasome activation, giving rise to chronic inflammation [17]. Multiple recent findings also provide evidence for the persistence of SARS-CoV-2 RNA and antigen as potential culprits for chronic low-grade systemic inflammation in PCS [18,19].

Studies presented by **Leonard A. Jason** provide insights into potential predisposing factors for ME/CFS and PCS via analyses of immune profiles prior to infection with Epstein-Barr-Virus (EBV) [20]. Data from 4501 healthy students were collected and followed up for the occurrence of ME/CFS. Among the participants, 5% of those who developed infectious mononucleosis (IM) were later diagnosed with ME/CFS. Individuals with deficiencies in Interleukin-5 (IL-5) and Interleukin-13 (IL-13) production were more likely to develop severe ME/CFS after EBV infection possibly due to immune dysregulation. Cytokine network analysis revealed more densely interconnected cytokine networks at least six weeks before IM in ME/CFS patients. Those with preceding severe gastrointestinal symptoms and low IL-5/IL-13 blood levels had a nearly 80% likelihood of developing severe ME/CFS. Moreover, ME/CFS patients showed dysregulated metabolic pathways essential for cell proliferation, particularly during pro-inflammatory immune responses [21]. Implications for the identification of predisposing factors can also be drawn from findings on PCS, where Jason and Dorri found cognitive and autonomic dysfunction to be predictors of SARS-CoV-2 infection associated with ME/CFS [21].

3. Diagnosing ME/CFS

Due to the complexity of symptoms, limited education and acceptance of the disease as well as poor access to medical care for severely affected patients, the diagnosis and treatment of ME/CFS can be difficult, and a high number of undiagnosed patients is presumed [22,23]. **Uta Behrends** presented the state-of-the-art diagnostic approach involving a comprehensive medical history, the evaluation of diagnostic criteria via appropriate questionnaires, physical examination, functional tests, routine laboratory analyses, and imaging as required for appropriate differential diagnostics. In the course of this, the newly developed Munich-Berlin Symptom Questionnaire (MBSQ) [24] was presented, which combines the CCC [14] and Institute of Medicine (IOM) criteria [25] and allows for a quantitation of symptoms. Such diagnostic assessments should be offered in specialized centers to achieve timely diagnosis and care for affected patients. Regular reassessments should be provided to evaluate the individual courses of disease, including partial or complete remissions, especially in children and adolescents [26,27].

Further lectures focused on the diagnosis and potential role of autonomous and skeletal muscle dysfunction, sleep disturbance, cognitive symptoms, and hypermobility. Symptoms of a dysfunctional autonomic nervous system in ME/CFS are complex and entail the involvement of the parasympathetic and sympathetic system. A sympathetic noradrenergic failure can manifest by orthostatic, exercise, and temperature intolerance. In contrast, patients with hyperactivity of the autonomic system show symptoms such as paleness, higher blood pressure, trembling, bristling hair, or sweating. **Pawel Zalewski** presented different phenotypes of autonomic dysfunction in ME/CFS, which emerged from a study of his research group [28]. Correct phenotyping is crucial in providing personalized therapy for affected individuals. This can be achieved through various examinations including blood pressure determination, monitoring of heart rate variability, and laboratory tests for neurotransmitters. Physical abilities should also be assessed through examinations like handgrip strength [29], and a NASA 10-min Lean Test. These tests serve as a basis for creating an individual treatment plan.

Physical and rehabilitation medicine can offer function-centered

diagnostics and therapy for ME/CFS patients, enabling an individual approach. **Max Liebl** presented findings of muscular dysfunction and abnormalities, with a focus on respiration, by conducting function-centered diagnostics in ME/CFS patients analyzed within the CFS_CARE study of the Charité [30]. Methods used for this purpose include measuring thoracic respiratory excursion and conducting manual diaphragm as well as cervical and thoracic spine examinations. These diagnostic tools provide a basis for individual physical therapy. Reflective respiratory therapy, which is a combination of thermal stimuli with special massage grip techniques for increased thoracic flexibility and harmonized breathing patterns, was particularly highlighted in this context [31]. Furthermore, the importance of home exercises for ME/CFS and PCS patients combining mobility and breathing exercises with self-applied relaxation strategies was emphasized.

In the cognition and magnetic resonance imaging (MRI) in post-COVID (CAMINO) study, characteristics of white matter and subcortical brain structures in patients with PCS-associated fatigue and cognitive impairment were examined [32]. **Carsten Finke** presented the results of this study unveiling that PCS is linked with deficits of attention, memory (learning, long-term, and working memory), and executive function (semantic fluency, dual-task). Structural MRI showed a reduced volume and impaired microstructural integrity of basal ganglia and the thalamus in association with PCS fatigue [32]. Analyses of a population-based sample of the German National Pandemic Cohort Network (NAPKON) study COVIDOM identified older age, shorter education, male gender, and history of neuropsychiatric disease as predictors for cognitive deficits as well as younger age, female gender, number of acute COVID symptoms and depression as predictors of fatigue in PCS [33]. Similar studies are ongoing in ME/CFS patients within the National Clinical Study Group (NKSJ) an interdisciplinary network of physicians and scientists to develop translational research and therapeutic trials for the treatment of ME/CFS and PCS [34].

One in three persons in Germany describes sleep disturbances, but only every fifth suffers from sleep disturbances at least three nights per week [35]. Obstructive sleep apnea (OSA) is more frequent in men than in women and its prevalence increases with age [36]. Sleep disturbances are described as obligatory ME/CFS symptom in current case definitions as defined by the IOM criteria [25], the CCC [14], and the National Institute for Health and Care Excellence (NICE) guidelines [37]. The CCC recommends the exclusion of restless legs syndrome and OSA in every CFS patient [14]. Affected people diagnosed within the ongoing CFS_CARE study at Charité [30], revealed a high prevalence of at least one sleep disorder (60 of 64 ME/CFS patients) as presented by **Christian Veauthier**. Hence, sleep disorders seem to be very common in ME/CFS and physicians are advised not to overlook them. Therefore, home sleep testing should be performed on every ME/CFS patient. Diagnosing and treating them according to official guidelines could result in improvement of fatigue and cognitive symptoms.

Peter Rowe highlighted the importance of diagnosing joint hypermobility (JH) and hypermobile Ehlers-Danlos syndrome (hEDS) as potential comorbidities of ME/CFS with, an increased prevalence in ME/CFS patients, as shown by various studies [38–40]. Often, hypermobility is accompanied by orthostatic intolerance (OI). Mechanisms of the association between OI and JH/hEDS are not fully understood, but there is evidence for connective tissue laxity in blood vessels [38], peripheral neuropathy [41], and mast cell activation syndrome [42]. Affected ME/CFS patients can benefit from the diagnosis of hypermobility syndromes in terms of appropriate clinical management [43].

4. Understanding ME/CFS

4.1. Endothelial dysfunction

Impaired blood flow and endothelial biomarkers were an important topic at the meeting. **Francisco Westermeier** elaborated on the importance of nitric oxide (NO) and the NO-forming enzyme endothelial

nitric oxide synthase (eNOS) for cardiovascular tissue and endothelial homeostasis. In the presented work conducted by Blauensteiner et al. [44], the Sirtuin 1 (SIRT1) – eNOS axis in ME/CFS was investigated and revealed an increased level of selected microRNAs associated with endothelial dysfunction. Additionally, Francisco Westermeier showed data that indicated an inhibitory phosphorylation effect of eNOS, which leads to a reduced NO production of endothelial cells (EC) incubated with plasma from ME/CFS patients [45]. Furthermore, preliminary data on L-arginine metabolites, which are essential for NO production, hint towards dysregulation in ME/CFS patients, with contrary patterns in males and females.

Martina Seifert investigated the functional effects of serum factors on ECs in vitro from PCS patients of which a subset had ME/CFS. Differences in the secretion of selected small molecules (e.g., Soluble Vascular Cell Adhesion Molecule-1 (sVCAM-1), Neutrophil Gelatinase-Associated Lipocalin (NGAL), Myeloperoxidase (MPO), myeloid-related protein (MRP) 8/14, and others) were found between PCS and PCS ME/CFS patients and healthy individuals after serum treatment of human EC [46], which may be related to a different pro-angiogenic potential. Interestingly, anti-EC-AABs (AECA) were more abundant in the sera of PCS/CFS patients [46]. Finally, plasma extracellular vesicles were discussed as potential biomarkers of altered endothelial function in PCS/CFS and ME/CFS, as preliminary data showed a markedly different phenotype and proteome compared with vesicles from healthy individuals.

As emphasized by **Christian Puta**, understanding PEM is crucial and requires analysis of exercise-induced responses. PEM is defined as an intolerance of mild to moderate mental and physical exertion with an aggravation of symptoms, which usually last longer than 14 h and can persist for several days or weeks with dysregulated acute exercise tolerance [47] and dysregulated recovery processes [2]. PCS and ME/CFS patients have a low aerobic capacity [16,17], and therefore anaerobic capacity should be considered in diagnostics. It was hypothesized that immunological and metabolic dysregulations, hypoxia, as well as dysfunctional microcirculation, and morphological changes in red blood cells leading to inadequate tissue perfusion might be causative for PEM. More generally, it is crucial that strategies to avoid PEM are considered in rehabilitation therapies for ME/CFS patients.

4.2. Immune dysregulation

The utilization of omics technologies has been instrumental in the characterization of immune dysregulation ongoing in severe acute COVID-19 [48] and led to the identification of substantial transcriptional deviations in immune cell subsets of the myeloid compartment and T cells [49]. **Anna Aschenbrenner** presented the first results from a study on a well-defined cohort of PCS patients including a subset fulfilling the CCC for ME/CFS using single-cell RNA sequencing for the analysis of circulating peripheral immune cells. In comparison to fully recovered healthy individuals 5–12 months after SARS-CoV-2 infection, those PCS patients showed a pro-inflammatory monocyte profile, pro-inflammatory NK cell subpopulations, and an increase in the number of cytotoxic CD16⁺ T cells.

Andreas Goebel presented research jointly conducted with Camilla Svensson (Karolinska Institute), David Andersson, and Stuart Bevan (Kings College London) [50] on findings of AABs to satellite glia cells (SGC). Injection of isolated immunoglobulin G (IgG) fractions from fibromyalgia syndrome (FMS) patients led to reduced locomotion, decreased skin innervation as well as hypersensitivities against mechanical and cold stimuli in mice. The binding of IgGs to SGC and neurons could be shown. Hence, anti-SGC AABs might explain the spontaneous pain described as a symptom in FMS patients. In preliminary studies on PCS, there was no clear evidence that pain was associated with anti-SGC AABs. Future efforts are needed to better understand whether PCS-associated pain can be passively transferred to mice.

Bettina Hohberger presented the impact of functional AAB (fAAB) targeting GPCR (GPCR-fAAB) on capillary microcirculation in patients with PCS, quantified in the macula by optical coherence tomography angiography (OCT-A) [51,52]. Hohberger reported on an experimental therapy of a patient with PCS and glaucoma, being seropositive for GPCR-fAABs, who was treated with the DNA aptamer BC007 for neutralization of the GPCR-fAABs. After this experimental treatment, the patient's PCS symptoms improved. This was accompanied by an improvement in capillary macular microcirculation, measured by OCT-A, and a decrease in intraocular pressure. Using a cardiomyocyte beat rate test system several GPCR-fAABs were observed in sera of patients with PCS, or glaucoma [53,54].

4.3. Viral reactivation

Nuno Sepúlveda presented a re-analysis of published data on IgG responses to over 3000 peptides derived from 14 EBV proteins in 92 ME/CFS patients and 50 healthy controls [55]. The study had the objective of investigating the potential role of EBV mimicry in triggering autoimmunity. A machine learning approach revealed stronger antibody responses against two peptides from EBNA4 and EBNA6 in ME/CFS patients with a putative infection in their disease onset compared to controls indicated that using these antibody responses together with age and gender the final classification model had good diagnostic sensitivity and specificity [56]. In a consecutive study, two peptides derived from EBNA6 were identified with high sequence homologies with different human proteins encoding by the following genes CCCTC-binding factor (CTCF, a master genomic regulator), adipocyte enhancer-binding protein 1 (AEBP1, associated with Ehlers-Danlos Syndrome), homeobox A9 (HOXA-9), and adrenergic receptor alpha 1B. Strikingly, these human genes and the respective coding proteins may play a role in ME/CFS pathology, via a putative EBV molecular mimicry.

The presentation given by **Bhupesh Prusty** dealt with the coherency of viral antibodies, autoimmunity, and mitochondrial dysfunction in ME/CFS pathogenesis. Patients with ME/CFS, PCS, and/or post-COVID fatigue showed high antibody responses against the deoxyuridine triphosphatase (dUTPase) of EBV, herpes simplex virus 1 (HSV-1), and human herpes virus type 6 (HHV-6), indicating a possible role of herpesvirus reactivation. Evidently, these specific dUTPases can induce mitochondrial damage. The second focus of the lecture was on a protein microarray analysis, which revealed a correlation between IgM antibodies (e.g., house dust mites, cat/dog hair) and ME/CFS disease severity, suggesting increased responsiveness to foreign antigens. Furthermore, ME/CFS patients in this study exhibit higher circulating fibronectin levels compared to controls, which may be associated with chronic inflammation and disruption of clot homeostasis. Additionally, presented data indicate that IgG fractions from patients with severe ME/CFS can induce mitochondrial fragmentation in ECs in vitro. These findings suggest that virus reactivation in localized tissues may contribute to mitochondrial dysfunction and tissue inflammation in ME/CFS.

5. Treatment of ME/CFS

5.1. State of the art and off-label therapies

Luis Nacul introduced the third theme of the conference, which addressed the treatment options for ME/CFS currently applied in practice and under clinical investigation. Patient care, including pharmacological and non-pharmacological treatment options, should be based on consented guidelines like the NICE guideline on ME/CFS (NG206) or the Expert Consensus of the European Network on ME/CFS (EURO-MENE) [6,37]. Specific treatment options should be agreed upon patient and health professional, following transparent discussions concerning existing evidence, the balance between potential benefits and risks, and patient preferences. Non-pharmacological interventions, like pacing,

highly personalized and specialized outpatient care as well as further aid in reducing symptom burden can improve the long-term prognosis of ME/CFS [6,57,58]. It is important to consider opportunities for early recognition and treatment of patients at all levels of health services. Training and education of health professionals, including in primary care, are important to enable better coverage and care of patients, particularly at early stages of the disease, when therapeutic interventions will likely be more beneficial [59].

Based on his broad clinical experience, **Michael Stingl** presented a range of neuromodulating drugs that can be helpful in ME/CFS. Short-time usage of benzodiazepines can be beneficial in lowering the symptom burden of PEM, sleep disturbances, or sensory overload due to their sedative/anxiolytic properties [60]. Anticonvulsants can suppress the excessive firing of neurons, reduce neuropathic pain, and counteract potential neuroinflammation [61]. Furthermore, antidepressants and antipsychotics with anti-inflammatory and neuromodulator properties can be administered to improve cognition and endothelial function [62–70]. Based on first clinical studies cholinesterase inhibitor pyridostigmine, used for myasthenia gravis therapy, is promising for treating vascular dysfunction, hypoperfusion, and muscle fatigue in ME/CFS [71]. OI in ME/CFS may manifest as POTS, orthostatic hypotension (OH), or even postural symptoms without tachycardia (PSWT). Non-invasive interventions for the treatment of all forms of OI are centered on identifying and avoiding individual triggers of symptom exacerbation (e.g. volume depletion, long standing, heat exposure), applying compression garments, pacing when strengthening leg and abdominal muscles, and increasing fluid and sodium intake [72]. Additionally to the non-pharmacological basis therapy, pharmacological treatments for POTS and OH entail a range of centrally and peripherally acting substances, including ivabradine, propranolol, midodrine, pyridostigmine, and fludrocortisone as presented by **Andrea Maier** and **Luis Nacul** [67,69,73]. Initial low-dose administration is not only recommended in patients with OI to provide better tolerability, but possible side effects have to be carefully monitored.

Johannes-Peter Haas introduced an interdisciplinary, multi-professional, multimodal approach for young patients with ME/CFS at the German Center for Pediatric and Adolescent Rheumatology in Garmisch-Partenkirchen. The main goals are to increase patients' ability to apply individual pacing efforts and to provide knowledge on how to improve blood circulation, implement sleep hygiene, and manage pain. The components of the therapy are fixed within a weekly schedule not exceeding 30 min each, whilst allowing for individual adaptations on a daily basis. Follow-up data after four months suggest lasting improvements in certain domains, including physical functioning.

Laura Froehlich presented a survey about the medical care situation in ME/CFS in Germany indicating that patients were severely medically underserved. Their overall satisfaction with the care provided by general practitioners was low and only one-third were being treated by physicians specializing in ME/CFS [74]. Findings from the same survey also point towards a high degree of stigmatization perceived by ME/CFS patients, resulting probably in lower functional status and satisfaction with social roles [75]. **Bettina Grande** emphasized that although psychotherapy and psychosomatic rehabilitation have no proven curative effect in ME/CFS, there is an important role for psychotherapeutic support. Whilst acknowledging the limitations of psychotherapy in the care of ME/CFS patients, psychotherapists can improve patients' ability to handle their individual energy levels, loneliness, and frustration to improve general well-being [76,77].

5.2. Clinical trials

Results of ongoing clinical trials and current perspectives on targeting endothelial dysfunction and hypoperfusion were presented. **Elisa Stein** and **Wolfgang Ries** both showed results of clinical trials on immunoadsorption, which had previously shown to be effective in subgroups of ME/CFS patients [78,79]. **Elisa Stein** presented the

interim results from an observational trial of immunoadsorption in patients with SARS-CoV-2-triggered ME/CFS. The study aims to investigate the effectiveness in improving physical functioning and reducing symptom burden, as well as tolerability of the treatment [80]. Preliminary data provide first evidence of efficacy in two-thirds of the patients. Effective depletion of total IgG, IgA, IgM, and anti- β 2 adrenergic receptor AAb levels was shown. **Wolfgang Ries** pointed out that immunoadsorption is a feasible treatment approach with good efficacy in severely affected ME/CFS patients. However, individualized measures have to be taken in order to prevent PEM, including specialized transport and the reduction of audio-visual stimuli. Preliminary data from 31 severely affected PCS and ME/CFS patients indicate a response rate of 70% after five treatment sessions.

Oystein Fluge provided an overview of the clinical trials investigating the B cell-depleting monoclonal antibody Rituximab (RituxME) and the alkylating cytotoxic agent Cyclophosphamide (CycloME) in ME/CFS [81,82]. Preliminary findings from a six-year follow-up indicated a more beneficial outcome in patients enrolled in the CycloME trial, compared to patients in the RituxME trial. Fluge et al. hypothesized that ongoing immune responses in ME/CFS may be characterized by a persisting pattern of IgG AAbs that emerge after systemic infection. Those antibodies originate from mature plasma cells (PC) and possibly target ECs, neurons in the autonomous nervous system as well as GPCRs, resulting in disturbed vascular function and blood flow regulation [83]. Since Rituximab showed limited efficacy possibly due to its inability to target mature PC, and considering the cytotoxicity of Cyclophosphamide, a small trial is currently investigating the feasibility of the anti-CD38 antibody Daratumumab targeting long-lived PC, and first evidence for efficacy was shown [84].

Finally, **Klaus Wirth** presented the mechanisms possibly underlying the disturbed balance of vasoconstriction and vasodilation, which results in hypoperfusion and impaired mitochondrial function. This might be triggered by AAbs targeting e.g., anti- β 2 adrenergic receptor, stress responses, or vascular damage following infection [85,86]. Potential treatment options should focus on the preferential vasodilation of blood vessels in the brain via soluble guanylate cyclase stimulators, phosphodiesterase type 5 (PDE5) inhibitors, autoantibody targeting, and stimulators of endothelial acetylcholine receptors [71,87–89]. Moreover, further novel therapeutics could direct the preferential vasodilation of skeletal muscle blood vessels.

6. Conclusions

In summary, recent advances in ME/CFS research, diagnosis, and treatment were discussed at this two-day meeting. Several innovative diagnostic and therapeutic approaches provided promising concepts of care for ME/CFS patients. The importance of translating research findings into therapeutic strategies was highlighted to improve the clinical outcomes of ME/CFS patients.

Authorship

SS, AF, FH, and SSc wrote the manuscript draft. All other authors made substantial scientific contributions during the conference by presenting and discussing their research, which is now summarized here in this report. Moreover, all authors reviewed the manuscript critically for important intellectual content and have given final approval for the version to be submitted.

Submission declaration

This work has not been previously published and is not under consideration for publication elsewhere. If accepted, this work will not be published elsewhere in the same form, in English, or any other language, including electronically without the written consent of the copyright holder.

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Declaration of Generative AI and AI-assisted technologies in the writing process

During the preparation of this work, the authors did not use generative AI and AI-assisted technologies in the writing process, which go beyond improving readability and language.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Data availability

No data was used for the research described in the article.

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References

- [1] Friedman KJ, Murovska M, Pheby DFH, Zalewski P. Our evolving understanding of ME/CFS. *Medicina* (Kaunas) 2021;57(3). <https://doi.org/10.3390/medicina57030200>.
- [2] Kedor C, Freitag H, Meyer-Arndt L, Wittke K, Hanitsch LG, Zoller T, et al. A prospective observational study of post-COVID-19 chronic fatigue syndrome following the first pandemic wave in Germany and biomarkers associated with symptom severity. *Nat Commun* 2022;13(1):5104. <https://doi.org/10.1038/s41467-022-32507-6>.
- [3] Legler F, Meyer-Arndt L, Mödl L, Kedor C, Freitag H, Stein E, et al. Long-term symptom severity and clinical biomarkers in post-COVID-19/chronic fatigue syndrome: results from a prospective observational cohort. *eClinicalMedicine*. 2023;63. <https://doi.org/10.1016/j.eclinm.2023.102146>.
- [4] Salari N, Khodayari Y, Hosseini-Far A, Zarei H, Rasoulpoor S, Akbari H, et al. Global prevalence of chronic fatigue syndrome among long COVID-19 patients: a systematic review and meta-analysis. *Biopsychosoc Med* 2022;16(1):21. <https://doi.org/10.1186/s13030-022-00250-5>.
- [5] Roessler M, Tesch F, Batram M, Jacob J, Loser F, Weidinger O, et al. Post-COVID-19-associated morbidity in children, adolescents, and adults: a matched cohort study including more than 157,000 individuals with COVID-19 in Germany. *PLoS Med* 2022;19(11):e1004122. <https://doi.org/10.1371/journal.pmed.1004122>.
- [6] Nacul L, Authier FJ, Scheibenbogen C, Lorusso L, Helland IB, Martin JA, et al. European network on Myalgic encephalomyelitis/chronic fatigue syndrome (EUROMENE): expert consensus on the diagnosis, service provision, and Care of People with ME/CFS in Europe. *Medicina*. 2021;57(5):510. <https://doi.org/10.3390/medicina57050510>.
- [7] Sotzny F, Blanco J, Capelli E, Castro-Marrero J, Steiner S, Murovska M, et al. Myalgic encephalomyelitis/chronic fatigue syndrome - evidence for an autoimmune disease. *Autoimmun Rev* 2018;17(6):601–9. <https://doi.org/10.1016/j.autrev.2018.01.009>.
- [8] Davis HE, McCorkell L, Vogel JM, Topol EJ. Long COVID: major findings, mechanisms and recommendations. *Nat Rev Microbiol* 2023;21(3):133–46. <https://doi.org/10.1038/s41579-022-00846-2>.
- [9] Haffke M, Freitag H, Rudolf G, Seifert M, Doehner W, Scherbakov N, et al. Endothelial dysfunction and altered endothelial biomarkers in patients with post-COVID-19 syndrome and chronic fatigue syndrome (ME/CFS). *J Transl Med* 2022;20(1):138. <https://doi.org/10.1186/s12967-022-03346-2>.
- [10] Ahamed J, Laurence J. Long COVID endotheliopathy: hypothesized mechanisms and potential therapeutic approaches. *J Clin Invest* 2022;132(15). <https://doi.org/10.1172/JCI161167>.
- [11] Cabral-Marques O, Marques A, Gill LM, De Vito R, Rademacher J, Gunther J, et al. GPCR-specific autoantibody signatures are associated with physiological and pathological immune homeostasis. *Nat Commun* 2018;9(1):5224. <https://doi.org/10.1038/s41467-018-07598-9>.
- [12] Ryabkova VA, Gavrilova NY, Fedotkina TV, Churilov LP, Shoenfeld Y. Myalgic encephalomyelitis/chronic fatigue syndrome and post-COVID syndrome: a common neuroimmune ground? *Diagnostics* (Basel) 2022;13(1). <https://doi.org/10.3390/diagnostics13010066>.
- [13] Ryabkova VA, Gavrilova NY, Poletaeva AA, Pukhalenko AI, Koshkina IA, Churilov LP, et al. Autoantibody correlation signatures in fibromyalgia and myalgic encephalomyelitis/chronic fatigue syndrome: association with symptom severity. *Biomedicines*. 2023;11(2). <https://doi.org/10.3390/biomedicines11020257>.
- [14] Carruthers BM, Jain AK, De Meirleir KL, Peterson DL, Klimas NG, Lerner AM, et al. Myalgic encephalomyelitis/chronic fatigue syndrome. *J Chronic Fat Syndr* 2003;11(1):7–115. https://doi.org/10.1300/J092v11n01_02.
- [15] Sotzny F, Filgueiras IS, Kedor C, Freitag H, Wittke K, Bauer S, et al. Dysregulated autoantibodies targeting vaso- and immunoregulatory receptors in post COVID syndrome correlate with symptom severity. *Front Immunol* 2022;13. <https://doi.org/10.3389/fimmu.2022.981532>.
- [16] Komaroff AL, Lipkin WI. ME/CFS and long COVID share similar symptoms and biological abnormalities: road map to the literature. *Front Med* 2023;10. <https://doi.org/10.3389/fmed.2023.1187163>.
- [17] Paul BD, Lemle MD, Komaroff AL, Snyder SH. Redox imbalance links COVID-19 and myalgic encephalomyelitis/chronic fatigue syndrome. *Proc Natl Acad Sci* 2021;118(34):e2024358118. <https://doi.org/10.1073/pnas.2024358118>.
- [18] Stein SR, Ramelli SC, Grazioli A, Chung J-Y, Singh M, Yinda CK, et al. SARS-CoV-2 infection and persistence in the human body and brain at autopsy. *Nature*. 2022;612(7941):758–63. <https://doi.org/10.1038/s41586-022-05542-y>.
- [19] Swank Z, Senussi Y, Manickas-Hill Z, Yu XG, Li JZ, Alter G, et al. Persistent circulating severe acute respiratory syndrome coronavirus 2 spike is associated with post-acute coronavirus disease 2019 sequelae. *Clin Infect Dis* 2022;76(3). <https://doi.org/10.1093/cid/ciac722>. e487–e90.
- [20] Jason LA, Cotler J, Islam MF, Sunnquist M, Katz BZ. Risks for developing Myalgic encephalomyelitis/chronic fatigue syndrome in college students following infectious mononucleosis: a prospective cohort study. *Clin Infect Dis* 2020;73(11). <https://doi.org/10.1093/cid/ciaa1886>. e3740–e6.
- [21] Jason LA, Cotler J, Islam MF, Furst J, Katz BZ. Predictors for developing severe myalgic encephalomyelitis/chronic fatigue syndrome following infectious mononucleosis. *J Rehabil Ther* 2022;4(1):1–5. <https://doi.org/10.29245/2767-5122/2021.1.1129>.
- [22] Jason LA, Katz BZ, Sunnquist M, Torres C, Cotler J, Bhatia S. The prevalence of pediatric Myalgic encephalomyelitis/chronic fatigue syndrome in a community-based sample. *Child Youth Care Forum* 2020;49(4):563–79. <https://doi.org/10.1007/s10566-019-09543-3>.
- [23] Araja D, Berkis U, Lunga A, Murovska M. Shadow burden of undiagnosed myalgic encephalomyelitis/chronic fatigue syndrome (ME/CFS) on society: Retrospective and prospective in light of COVID-19. *J Clin Med* 2021;10(14). <https://doi.org/10.3390/jcm10143017>.
- [24] Peo LC, Wiehler K, Paulick J, Gerrer K, Leone A, Viereck A, et al. Pediatric and adult patients with ME/CFS following COVID-19: A structured approach to diagnosis using the Munich Berlin Symptom Questionnaire (MBSQ). *medRxiv*. 2023. <https://doi.org/10.1101/2023.08.23.23293081>.

- [25] Committee on the Diagnostic Criteria for Myalgic Encephalomyelitis/Chronic Fatigue Syndrome, Board on the Health of Select Populations IoM. The National Academies Collection: Reports funded by National Institutes of Health. Beyond Myalgic Encephalomyelitis/Chronic Fatigue Syndrome: Redefining an Illness. Washington (DC): National Academies Press (US); 2015 (Copyright 2015 by the National Academy of Sciences. All rights reserved).
- [26] Rowe KS. Long term follow up of young people with chronic fatigue syndrome attending a pediatric outpatient service. *Front Pediatr* 2019;7. <https://doi.org/10.3389/fped.2019.00021>.
- [27] Pricoco R, Meidel P, Hofberger T, Zietemann H, Mueller Y, Wiehler K, et al. One-year follow-up of young people with ME/CFS following infectious mononucleosis by Epstein-Barr virus. *medRxiv* 2023. <https://doi.org/10.1101/2023.07.24.23293082>.
- [28] Stomko J, Estévez-López F, Kujawski S, Zawadka-Kunikowska M, Tafil-Klawe M, Klawe JJ, et al. Autonomic phenotypes in Chronic Fatigue Syndrome (CFS) are associated with illness severity: A cluster analysis. *J Clin Med* 2020;9(8). <https://doi.org/10.3390/jcm9082531>.
- [29] Jäkel B, Kedor C, Grabowski P, Wittke K, Thiel S, Scherbakov N, et al. Hand grip strength and fatigability: correlation with clinical parameters and diagnostic suitability in ME/CFS. *J Transl Med* 2021;19(1):1–12. <https://doi.org/10.1186/s12967-021-02774-w>.
- [30] Charite University B, Germany. CFS CARE - Care Model for Patients with Chronic Fatigue Syndrome (ME/CFS) [Clinical Trial] [Available from:]; https://cfc.charite.de/klinische_studien/cfs_care/; 2023.
- [31] Best N, Seidel E. Consensus conference 2021/2022: physical and rehabilitative medicine – diagnostic and therapeutic options. *Physikalische Medizin, Rehabilitationsmed Kurortmed* 2022;32(02):73–81. <https://doi.org/10.1055/a-1767-0652>.
- [32] Heine J, Schwichtenberg K, Hartung TJ, Rekers S, Chien C, Boesl F, et al. Structural brain changes in patients with post-COVID fatigue: a prospective observational study. *EClinicalMedicine*. 2023;58:101874. <https://doi.org/10.1016/j.eclinm.2023.101874>.
- [33] Hartung TJ, Neumann C, Bahmer T, Chaplinskaya-Sobol I, Endres M, Geritz J, et al. Fatigue and cognitive impairment after COVID-19: a prospective multicentre study. *EClinicalMedicine*. 2022;53:101651. <https://doi.org/10.1016/j.eclinm.2022.101651>.
- [34] Scheibenbogen C, Bellmann-Strobl JT, Heindrich C, Wittke K, Stein E, Franke C, et al. Fighting post-COVID and ME/CFS – development of curative therapies. *Front Med* 2023;10. <https://doi.org/10.3389/fmed.2023.1194754>.
- [35] Ohayon MM. Epidemiology of insomnia: what we know and what we still need to learn. *Sleep Med Rev* 2002;6(2):97–111. <https://doi.org/10.1053/smr.2002.0186>.
- [36] Fietze I, Laharnar N, Obst A, Ewert R, Felix SB, Garcia C, et al. Prevalence and association analysis of obstructive sleep apnea with gender and age differences - results of SHIP-trend. *J Sleep Res* 2019;28(5):e12770. <https://doi.org/10.1111/jsr.12770>.
- [37] National Institute for Health and Care Excellence: Guidelines. Myalgic encephalomyelitis (or encephalopathy)/chronic fatigue syndrome: diagnosis and management. London: National Institute for Health and Care Excellence (NICE); 2021.
- [38] Rowe PC, Barron DF, Calkins H, Maumenee IH, Tong PY, Geraghty MT. Orthostatic intolerance and chronic fatigue syndrome associated with Ehlers-Danlos syndrome. *J Pediatr* 1999;135(4):494–9. [https://doi.org/10.1016/s0022-3476\(99\)70173-3](https://doi.org/10.1016/s0022-3476(99)70173-3).
- [39] Nijs J, Aerts A, De Meirleir K. Generalized joint hypermobility is more common in chronic fatigue syndrome than in healthy control subjects. *J Manipulative Physiol Ther* 2006;29(1):32–9. <https://doi.org/10.1016/j.jmpt.2005.11.004>.
- [40] Eccles JA, Thompson B, Themelis K, Amato ML, Stocks R, Pound A, et al. Beyond bones: the relevance of variants of connective tissue (hypermobility) to fibromyalgia, ME/CFS and controversies surrounding diagnostic classification: an observational study. *Clin Med (Lond)* 2021;21(1):53–8. <https://doi.org/10.7861/clinmed.2020-0743>.
- [41] Gazit Y, Nahir AM, Grahame R, Jacob G. Dysautonomia in the joint hypermobility syndrome. *Am J Med* 2003;115(1):33–40. [https://doi.org/10.1016/s0002-9343\(03\)00235-3](https://doi.org/10.1016/s0002-9343(03)00235-3).
- [42] Kohn A, Chang C. The relationship between hypermobile Ehlers-Danlos syndrome (hEDS), postural orthostatic tachycardia syndrome (POTS), and mast cell activation syndrome (MCAS). *Clin Rev Allergy Immunol* 2020;58(3):273–97. <https://doi.org/10.1007/s12016-019-08755-8>.
- [43] Roma M, Marden CL, De Wande I, Francomano CA, Rowe PC. Postural tachycardia syndrome and other forms of orthostatic intolerance in Ehlers-Danlos syndrome. *Auton Neurosci* 2018;215:89–96. <https://doi.org/10.1016/j.autneu.2018.02.006>.
- [44] Blauensteiner J, Bertinat R, León LE, Riederer M, Sepúlveda N, Westermeier F. Altered endothelial dysfunction-related miRs in plasma from ME/CFS patients. *Sci Rep* 2021;11(1):10604. <https://doi.org/10.1038/s41598-021-89834-9>.
- [45] Bertinat R, Villalobos-Labra R, Hofmann L, Blauensteiner J, Sepúlveda N, Westermeier F. Decreased NO production in endothelial cells exposed to plasma from ME/CFS patients. *Vasc Pharmacol* 2022;143:106953. <https://doi.org/10.1016/j.vph.2022.106953>.
- [46] Flaskamp L, Roubal C, Uddin S, Sotzny F, Kedor C, Bauer S, et al. Serum of post-COVID-19 syndrome patients with or without ME/CFS differentially affects endothelial cell function in vitro. *Cells* 2022;11(15). <https://doi.org/10.3390/cells11152376>.
- [47] Singh I, Leitner BP, Wang Y, Zhang H, Joseph P, Lutchmansingh DD, et al. Proteomic profiling demonstrates inflammatory and endotheliopathy signatures associated with impaired cardiopulmonary exercise hemodynamic profile in post acute sequelae of SARS-CoV-2 infection (PASC) syndrome. *Pulm Circ* 2023;13(2):e12220. <https://doi.org/10.1002/pul2.12220>.
- [48] Schultze JL, Aschenbrenner AC. COVID-19 and the human innate immune system. *Cell*. 2021;184(7):1671–92. <https://doi.org/10.1016/j.cell.2021.02.029>.
- [49] Georg P, Astaburuaga-García R, Bonaguro L, Brumhard S, Michalick L, Lippert LJ, et al. Complement activation induces excessive T cell cytotoxicity in severe COVID-19. *Cell* 2022;185(3). <https://doi.org/10.1016/j.cell.2021.12.040>. 493–512.e25.
- [50] Goebel A, Krock E, Gentry C, Israel MR, Jurczak A, Urbina CM, et al. Passive transfer of fibromyalgia symptoms from patients to mice. *J Clin Invest* 2021;131(13). <https://doi.org/10.1172/JCI144201>.
- [51] Szewczykowski C, Mardin C, Lucio M, Wallukat G, Hoffmanns J, Schröder T, et al. Long COVID: Association of functional autoantibodies against G-Protein-Coupled receptors with an impaired retinal microcirculation. *Int J Mol Sci* 2022;23(13). <https://doi.org/10.3390/jms23137209>.
- [52] Schlick S, Lucio M, Wallukat G, Bartsch A, Skornia A, Hoffmanns J, et al. Post-COVID-19 syndrome: Retinal microcirculation as a potential marker for chronic fatigue. *Int J Mol Sci* 2022;23(22). <https://doi.org/10.3390/jms232213683>.
- [53] Wallukat G, Hohberger B, Wenzel K, Fürst J, Schulze-Rothe S, Wallukat A, et al. Functional autoantibodies against G-protein coupled receptors in patients with persistent long-COVID-19 symptoms. *J Transl Autoimmun* 2021;4:100100. <https://doi.org/10.1016/j.jtauto.2021.100100>.
- [54] Hohberger B, Kunze R, Wallukat G, Kara K, Mardin CY, Lämmer R, et al. Autoantibodies activating the β 2-adrenergic receptor characterize patients with primary and secondary Glaucoma. *Front Immunol* 2019;10. <https://doi.org/10.3389/fimmu.2019.02112>.
- [55] Loebel M, Eckey M, Sotzny F, Hahn E, Bauer S, Grabowski P, et al. Serological profiling of the EBV immune response in chronic fatigue syndrome using a peptide microarray. *PLoS One* 2017;12(6):e0179124. <https://doi.org/10.1371/journal.pone.0179124>.
- [56] Sepúlveda N, Malato J, Sotzny F, Grabowska AD, Fonseca A, Cordeiro C, et al. Revisiting IgG antibody reactivity to Epstein-Barr virus in myalgic encephalomyelitis/chronic fatigue syndrome and its potential application to disease diagnosis. *Front Med (Lausanne)* 2022;9:921101. <https://doi.org/10.3389/fmed.2022.921101>.
- [57] Jason L, Benton M, Torres-Harding S, Muldowney K. The impact of energy modulation on physical functioning and fatigue severity among patients with ME/CFS. *Patient Educ Couns* 2009;77(2):237–41. <https://doi.org/10.1016/j.pec.2009.02.015>.
- [58] Magel T, Meagher E, Boulter T, Albert A, Tsai M, Muñoz C, et al. Fatigue presentation, severity, and related outcomes in a prospective cohort following post-COVID-19 hospitalization in British Columbia, Canada. *Front Med (Lausanne)* 2023;10:1179783. <https://doi.org/10.3389/fmed.2023.1179783>.
- [59] Nacul L, O'Boyle S, Palla L, Nacul FE, Mudie K, Kingdon CC, et al. How Myalgic encephalomyelitis/chronic fatigue syndrome (ME/CFS) progresses: the natural history of ME/CFS. *Front Neurol* 2020;11:826. <https://doi.org/10.3389/fneur.2020.00826>.
- [60] Carruthers BM, van de Sande MI, De Meirleir KL, Klimas NG, Broderick G, Mitchell T, et al. Myalgic Encephalomyelitis – Adult & Paediatric: International Consensus Primer for Medical Practitioners: Carruthers & van de Sande. 2012.
- [61] de Greef BTA, Hoeijmakers JGJ, Geerts M, Oakes M, Church TJE, Waxman SG, et al. Lacosamide in patients with Nav1.7 mutations-related small fibre neuropathy: a randomized controlled trial. *Brain*. 2019;142(2):263–75. <https://doi.org/10.1093/brain/awy329>.
- [62] Lopez-Vilchez I, Diaz-Ricart M, Navarro V, Torramade S, Zamorano-Leon J, Lopez-Farre A, et al. Endothelial damage in major depression patients is modulated by SSRI treatment, as demonstrated by circulating biomarkers and an in vitro cell model. *Transl Psychiatry* 2016;6(9). <https://doi.org/10.1038/tp.2016.156>. e886-e.
- [63] Hashimoto K. Overview of the potential use of fluvoxamine for COVID-19 and long COVID. *Discover Mental Health* 2023;3(1):9. <https://doi.org/10.1007/s44192-023-00036-3>.
- [64] Bennabi D, Haffen E, Van Waes V. Vortioxetine for cognitive enhancement in major depression: from animal models to clinical research. *Front Psychol* 2019;10. <https://doi.org/10.3389/fpsy.2019.00771>.
- [65] Talmon M, Rossi S, Pastore A, Cattaneo CI, Brunelleschi S, Fresu LG. Vortioxetine exerts anti-inflammatory and immunomodulatory effects on human monocytes/macrophages. *Br J Pharmacol* 2018;175(1):113–24. <https://doi.org/10.1111/bph.14074>.
- [66] Ślusarczyk J, Trojan E, Głombik K, Piotrowska A, Budziszewska B, Kubera M, et al. Targeting the NLRP3 Inflammasome-related pathways via Tianeptine treatment-suppressed microglia polarization to the M1 phenotype in lipopolysaccharide-stimulated cultures. *Int J Mol Sci* 2018;19(7):1965. <https://doi.org/10.3390/jms19071965>.
- [67] Crosby LD, Kalanidhi S, Bonilla A, Subramanian A, Ballon JS, Bonilla H. Off label use of aripiprazole shows promise as a treatment for Myalgic encephalomyelitis/chronic fatigue syndrome (ME/CFS): a retrospective study of 101 patients treated with a low dose of aripiprazole. *J Transl Med* 2021;19(1):50. <https://doi.org/10.1186/s12967-021-02721-9>.
- [68] Toljan K, Vrooman B. Low-dose naltrexone (LDN)—review of therapeutic utilization. *Med Sci* 2018;6(4):82. <https://doi.org/10.3390/medsci6040082>.
- [69] Polo O, Pesonen P, Tuominen E. Low-dose naltrexone in the treatment of myalgic encephalomyelitis/chronic fatigue syndrome (ME/CFS). *Fat: Biomed Health Behav* 2019;7(4):207–17. <https://doi.org/10.1080/21641846.2019.1692770>.
- [70] Jansen van Vuren E, Steyn SF, Brink CB, Möller M, Viljoen FP, Harvey BH. The neuropsychiatric manifestations of COVID-19: Interactions with psychiatric illness

- and pharmacological treatment. *Biomed Pharmacother* 2021;135:111200. <https://doi.org/10.1016/j.biopha.2020.111200>.
- [71] Joseph P, Pari R, Miller S, Warren A, Stovall MC, Squires J, et al. Neurovascular dysregulation and acute exercise intolerance in Myalgic encephalomyelitis/chronic fatigue syndrome: a randomized. Placebo-Controlled Trial Pyridostig CHEST 2022; 162(5):1116–26. <https://doi.org/10.1016/j.chest.2022.04.146>.
- [72] Raj SR, Guzman JC, Harvey P, Richer L, Schondorf R, Seifer C, et al. Canadian cardiovascular society position statement on postural orthostatic tachycardia syndrome (POTS) and related disorders of chronic orthostatic intolerance. *Can J Cardiol* 2020;36(3):357–72. <https://doi.org/10.1016/j.cjca.2019.12.024>.
- [73] Raj SR, Fedorowski A, Sheldon RS. Diagnosis and management of postural orthostatic tachycardia syndrome. *Cmaj*. 2022;194(10):E378–e85. <https://doi.org/10.1503/cmaj.211373>.
- [74] Froehlich L, Hattesoil DBR, Jason LA, Scheibenbogen C, Behrends U, Thoma M. Medical care situation of people with Myalgic encephalomyelitis/chronic fatigue syndrome in Germany. *Medicina*. 2021;57(7):646. <https://doi.org/10.3390/medicina57070646>.
- [75] Froehlich L, Hattesoil DB, Cotler J, Jason LA, Scheibenbogen C, Behrends U. Causal attributions and perceived stigma for myalgic encephalomyelitis/chronic fatigue syndrome. *J Health Psychol* 2022;27(10):2291–304. <https://doi.org/10.1177/13591053211027631>.
- [76] Grande T, Grande B, Gerner P, Hammer S, Stingl M, Vink M, et al. The role of psychotherapy in the Care of Patients with Myalgic encephalomyelitis/chronic fatigue syndrome. *Medicina*. 2023;59(4):719. <https://doi.org/10.3390/medicina59040719>.
- [77] O'Connor K, Sunnquist M, Nicholson L, Jason LA, Newton JL, Strand EB. Energy envelope maintenance among patients with myalgic encephalomyelitis and chronic fatigue syndrome: implications of limited energy reserves. *Chronic Illn* 2019;15(1): 51–60. <https://doi.org/10.1177/1742395317746470>.
- [78] Tolle M, Freitag H, Antelmann M, Hartwig J, Schuchardt M, van der Giet M, et al. Myalgic encephalomyelitis/chronic fatigue syndrome: Efficacy of repeat immunoadsorption. *J Clin Med* 2020;9(8). <https://doi.org/10.3390/jcm9082443>.
- [79] Scheibenbogen C, Loebel M, Freitag H, Krueger A, Bauer S, Antelmann M, et al. Immunoadsorption to remove ss2 adrenergic receptor antibodies in chronic fatigue syndrome CFS/ME. *PLoS One* 2018;13(3):e0193672. <https://doi.org/10.1371/journal.pone.0193672>.
- [80] Charite University B. Germany. Repeat Immunoadsorption Post Covid ME/CFS [Clinical Trial] [Available from:], <https://classic.clinicaltrials.gov/ct2/show/NCT05629988>; 2022.
- [81] Fluge Ø, Rekeland IG, Lien K, Thürmer H, Borchgrevink PC, Schäfer C, et al. B-lymphocyte depletion in patients with Myalgic encephalomyelitis/chronic fatigue syndrome. *Ann Intern Med* 2019;170(9):585–93. <https://doi.org/10.7326/M18-1451>.
- [82] Rekeland IG, Fosså A, Lande A, Ktoridou-Valen I, Sørland K, Holsen M, et al. Intravenous cyclophosphamide in myalgic encephalomyelitis/chronic fatigue syndrome. An open-label Phase II study. *Front Med* 2020;7. <https://doi.org/10.3389/fmed.2020.00162>.
- [83] Fluge Ø, Tronstad KJ, Mella O. Pathomechanisms and possible interventions in myalgic encephalomyelitis/chronic fatigue syndrome (ME/CFS). *J Clin Invest* 2021;131(14). <https://doi.org/10.1172/jci150377>.
- [84] Fluge Ø. Behandling med daratumumab injeksjoner ved myalgisk encefalomyelitt/kronisk utmattelsessyndrom (ME/CFS) - en pilotstudie [Clinical Trial] [Available from:], <https://app.cristin.no/projects/show.jsf?id=2538921>; 2023.
- [85] Wirth KJ, Scheibenbogen C. Pathophysiology of skeletal muscle disturbances in Myalgic encephalomyelitis/chronic fatigue syndrome (ME/CFS). *J Transl Med* 2021;19(1):162. <https://doi.org/10.1186/s12967-021-02833-2>.
- [86] Wirth K, Scheibenbogen C. A unifying hypothesis of the pathophysiology of Myalgic encephalomyelitis/chronic fatigue syndrome (ME/CFS): recognitions from the finding of autoantibodies against ss2-adrenergic receptors. *Autoimmun Rev* 2020;19(6):102527. <https://doi.org/10.1016/j.autrev.2020.102527>.
- [87] Leitzke M. Is the post-COVID-19 syndrome a severe impairment of acetylcholine-orchestrated neuromodulation that responds to nicotine administration? *Bioelectr Med* 2023;9(1):2. <https://doi.org/10.1186/s42234-023-00104-7>.
- [88] Tosato M, Calvani R, Picca A, Ciciarello F, Galluzzo V, Coelho-Júnior HJ, et al. Effects of L-arginine plus vitamin C supplementation on physical performance, endothelial function, and persistent fatigue in adults with long COVID: a single-blind randomized controlled trial. *Nutrients*. 2022;14(23):4984. <https://doi.org/10.3390/nu14234984>.
- [89] Charite University B. Germany; Bayer. Study to Investigate Improvement in Physical Function in SF-36 With Vericiguat Compared With Placebo in Participants With Post-COVID-19 Syndrome [Clinical Trial] [Available from:], <https://classic.clinicaltrials.gov/ct2/show/NCT05697640>; 2023.