An ME Association Research Summary: Reactivation of human herpesviruses and their role in ME/CFS and Long Covid

By Dr Katrina Pears 20 January 2023

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Introduction

In this research summary we look at the role of reactivated human herpesviruses (HHV) in ME/CFS and Long Covid. This is because a number of recent studies have implied that HHV's could be involved in disease pathology causing and perpetuating symptoms. And because this might lead to effective treatments and diagnostic tests.

HHVs have the ability to trick their host's immune system. This means it doesn't completely eradicate them and the virus can remain dormant (latent) only to be reactivated at a later stage. Latency helps the virus to survive and following a trigger, to spread. Once reactivated they can cause temporary symptoms or persistent disease. Latent viruses are triggered in multiple ways including by other infections, a weakened immune system, stress, trauma and by hormonal changes.

The onset of ME/CFS often follows a viral infection and the most common is Epstein Barr Virus (HHV4). Reactivated HHVs have been linked to ME/CFS and a number of other diseases, including Alzheimer's and Multiple Sclerosis. We

know that Covid-19 (a Coronavirus) is the trigger for Long Covid, but this virus might also cause dormant HHVs to become active and contribute to symptoms and chronic ill-health.

Background

There are over 100 different herpesviruses, but only 8 are commonly found to infect humans. These include HHV4, HHV6, HHV7, and HHV8.

The most common HHV is Epstein-Barr Virus (EBV) (or HHV4). It is found all over the world and has infected almost the entire population. Most people experience a mild infection during childhood, but infection in teenage years can develop into glandular fever, or mononucleosis, and is also known as 'the kissing disease'.

Another well-known and relatively common infection is Chickenpox (HHV3), caused by the Varicella-Zoster Virus. This is often an acute infection that is nearly always caught in childhood, with reactivation potentially causing Shingles in later life.

Cytomegalovirus (CMV) (HHV5) causes widespread infection, which is usually harmless, unless contracted during pregnancy where it can potentially harm babies, or by people with a weakened immune system where it can be fatal. In the USA nearly 1 in 3 children have been infected by CMV by the age of 5.

Previous Research

Until now, research in this area has yielded inconclusive results, for example:

- **Burbelo *et al.* 2012** found no serological evidence for HHV6 being involved in the pathogenesis of ME/CFS.
- **Cameron *et al.* 2010** also could not support the hypothesis of ongoing or reactivated EBV, HHV6 or CMV infection in the pathogenesis of ME/CFS.
- Studies investigating a biomarker (signature) of HHV for ME/CFS have also proved to be disappointing with much more work needed to prove a link: **Cox *et al.*, 2022**; **Sepúlveda *et al.*, 2022**.
- Other studies have looked at treating the reactivation of HHV6 and HHV7 using valaciclovir, valganciclovir, and artesunate with little success: **Maltsev, 2022**.

Despite these disappointing studies, promising results were found by, for example:

- **Loebel *et al.* 2014** showed it is likely that there is an impaired ability in ME/CS to control the early stages of EBV reactivation, owing to the lack of EBV-specific B- and T-cell's memory response.

- **Chapenko *et al.* 2012** discovered a high active viral load of HHV6, HHV7 and *Parvovirus B19* in people with ME/CFS compared to controls in a reasonably sized study. They also found an association between active viral infection and symptoms. Similarly, EBV has been shown to have a lasting immunological imprint with T-cell activation in **Fevang *et al.* 2021.**
- Fluctuating viral DNA loads of HHVs have also been shown to correlate with symptoms by **Lee *et al.* 2021** and to the severity of ME/CFS as shown by **Gravelsina *et al.* 2022.**
- Subgroups of ME/CFS patients with different infectious triggers have been found to be distinguished by herpesvirus serology (blood): **Domingues *et al.*, 2021**.
- Other studies have suggested a clinical benefit in a subset of patients with ME/CFS when treating reactivated HHVs with valganciclovir: **Montoya *et al.*, 2013**. And a study on those with central nervous system dysfunction and long-standing fatigue, with HHV6 and EBV infection, determined a resolution of symptoms with valganciclovir: **Kogelnik *et al.* 2006**.

Latest Research

1. **Apostolou E et al:**
   Saliva antibody-fingerprint of reactivated latent viruses after mild/asymptomatic COVID-19 is unique in patients with myalgic-encephalomyelitis/chronic fatigue syndrome
   Frontier in Immunology 13: 949787 | 20 October 2022

Background:
The researchers from Linköping University in Sweden carried out a novel piece of research on the reactivation of viruses after a mild or asymptomatic Covid-19 infection in ME/CFS. They analysed a range of viruses in the blood and saliva of 95 non-vaccinated ME/CFS patients and 110 healthy controls (referred to as healthy donors by the authors).

Results:
The results showed that following a Covid-19 infection, both groups had significant latent virus reactivation, with herpesviruses (EBV, HHV6) and endogenous retroviruses (HERV-K) being detected in the saliva.

However, in the ME/CFS patients this response was significantly stronger, in particular EBV-encoded nuclear antigen-1 (EBNA1) IgG. Results show that even mild or asymptomatic Covid-19 infection can be a potential trigger for these latent viruses, meaning that ME/CFS patients can be more susceptible and have a stronger reaction to a Covid-19 infection. This provides further evidence for an altered immune response in ME/CFS.

Insight:
Results from this study are interesting and the study has strength in its reasonable scale. Findings could explain why, when infected by other viruses,

ME/CFS patients tend to become more unwell and for longer than healthy controls, due to the role of dormant viruses being activated.

It is interesting that results were only significant in the saliva and not the blood, which could have implications in future studies, and lead to more significant results. The authors suggested this could be due to herpesviruses being common in the oral cavity making them easily traceable in saliva. It should also be noted that previous studies have shown that when studying antibodies for Covid-19, the saliva should be used.

2. **Kasimir F et al.**
   Tissue specific signature of HHV-6 infection in ME/CFS
   Frontiers in Molecular Biosciences 9:1044964 | 14 December 2022

Background:
Researchers based in Germany and the United States looked at the tissue-specific signature of human herpesvirus infection in ME/CFS, focusing on HHV-6 and EBV. This study analysed **post-mortem tissue** of 3 ME/CFS patients and 26 healthy controls.

Results:
The results revealed a high viral load in the brain and neuronal tissue, including the spinal cord in the ME/CFS samples, which was absent in the control group. Specifically, they found a high abundance of microRNA (molecules that help make proteins) from the HHVs.

The team hypothesised that this large amount of HHV microRNAs might lead to a decrease in mitochondrial function, which would change mitochondrial metabolism and the innate immune response (the immune defence you were born with). The authors suggested that the virus being found in the brain and neuronal tissue could affect nerve function and immune response as well as causing the characteristic symptoms experienced in ME/CFS.

The results suggest tissue-specific locations for the active viruses which might explain why a number of previous studies on HHV and EBV have provided disappointing results because of the mostly inaccessible sample locations.

Insight:
Results from this study are fascinating. Studies of this nature are incredibly rare and expanding the size of the ME/CFS cohort to verify the results will be very difficult, which is the main limitation of this research.

However, we sincerely thank those who donated the post-mortem tissue, members of the research team, and the Cambridge Brain Bank (with whom the ME Association has had a relationship for many years) for collecting and providing tissue samples.

We recognise the importance of developing an ME/CFS Post-Mortem Tissue Bank. A suitable location would be needed to collect and store tissue, which is currently being explored by the charity and members of the ME/CFS Biobank team. We issued a recent statement about this potential project following Kara Jane Spencer's tragic death and her ongoing appeal.

3. **Peluso et al.**
   Impact of pre-existing chronic viral infection and reactivation on the development of long COVID
   Journal of Clinical Investigation: e163669 | 22 July 2022

[E-pub. ahead of print]

Background:
Researchers from San Francisco investigated the presence and reactivation of chronic viral infections in Long Covid patients. They investigated EBV and CMV (which are HHVs) and HIV (Human Immunodeficiency Virus). The study analysed the blood of 280 adults who had had a Covid-19 infection and looked at the statistical associations between viral presence and Long Covid symptoms.

Results:
The study revealed that Long Covid symptoms were associated with evidence of recent EBV reactivation and pre-existing HIV infection (however, the statistical analysis was adjusted for participant factors, sample timing, comorbid conditions and prior hospitalization). Underlying CMV infection was associated with a decreased risk of Long Covid.

In summary, the results suggest differential effects of chronic viral co-infections on the likelihood of developing Long Covid and predicted distinct syndromic patterns.

Insight:
Results in this study are not conclusive. They only suggest that a recent EBV reactivation could lead to an increased risk of developing Long Covid, as Long Covid was found to have developed in those without EBV reactivation or CMV disease. It would seem, therefore, that viral reactivation is not essential to the development of Long Covid.

Without diving too deeply into the statistical analysis, we would question the extent to which the authors adjusted their data to get a positive statistical result and, in conclusion, would suggest that this is the weakest of the 3 recent studies in this area.

Conclusion

From the 3 studies reviewed above, we can conclude that:

- Fluctuating symptoms in ME/CFS could be a result of repeated HHV reactivation, and abnormal immune responses following a triggering event such as infection with another virus.
- Saliva is a good to use when studying HHV's due it being key to transmission, with the salivary gland being a particularly permissive site for HHV replication. But results also suggest the active virus may hide in inaccessible locations requiring post-mortem access.
- Results may vary depending on the HHV studied.

Existing research has shown some variation in results, especially as it examined different HHVs using different samples (blood, saliva, or post-mortem tissue). But we hope they are used as the basis for more research, as larger cohorts and more conclusive results could lead to the development of new and effective treatments that might boost antiviral immune responses and lead to immunological tests that improve the effectiveness of ME/CFS (and Long Covid) diagnosis.

New Research

Research is currently underway at Brunel University in London into the role of reactivated HHV-6B infection in ME/CFS and Long Covid. It is led by 2 members of the UK ME/CFS Biobank team: Professor Jackie Cliff at Brunel and Dr Eliana Lacerda at the London School of Hygiene and Tropical Medicine:

"We hypothesise that the characteristic persistent remitting and relapsing nature of the ME/CFS syndrome in many individuals is the result of repeated HHV-6B reactivation, due to abnormal immune responses in susceptible people following an initial triggering event such as infection with another virus."

It will focus on participants recording their symptom severity to determine whether changes in HHV-6B concentration in saliva occurs before or after changes in ME/CFS symptoms. This will allow scientists to determine if HHV-6B causes the symptom changes or if these occur after the disease gets worse.

The study will also investigate immune cell function, specifically in cytotoxic T cells, including responses to stimulation, and at the impact of HHV-6B on other immune cells. A pilot study from this team has already demonstrated that the concentration of DNA from HHV-6B was higher in saliva from people with ME/CFS, and that the concentration correlated with the severity of symptoms (Lee ***et al.*, 2021**).

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