Herpes Simplex Virus Type 1 and Alzheimer’s disease: Increasing Evidence for a major role of the virus
Introduction and Agenda
Agenda

- What is the HSV1 and why is it linked to AD?
- What is CMV and its relationship to AD?
- HSV1 reactivation in human CNS
- Effects of HSV1 neural infections
- HSV1 infected cell cultures
- Epidemiological Studies
- Genetic Studies
- CMV vs. HSV1: which one is the bigger player in AD?
- Possible antiviral treatments
HSV1 Virus

An introduction
What is HSV1?

- Estimated 3.7 billion people under the age of 50, or 67% of the population, had HSV-1 infection.
- Once infected, your body retains the virus for life
- The link between Herpes Simplex Virus Type 1 and Alzheimer’s Disease patho-immunology was introduced several decades ago
- HSV1 present in APOE-4 carrier brains plays a major role in A.D
  - 60% increased risk
- Suggested that HSV1 virus enters the brain in older age as result of decline of immune system
HSV1 in the body

- HSV1 periodically reactivates in the brain during episodes of stress, immunosuppression or inflammation
- Typically appears as cold sores around the mouth although some people are asymptomatic
- The virus has the ability to cause accumulation of beta amyloid and AD like TAU (p-TAU)
  - Aβ is an innate immune protein that protects against fungal and bacterial infections
Apolipoprotein-E (APOE)

- A protein involved in the transport of lipids that is strongly linked to AD pathogenesis
- The type 4 allele is present in approximately 10-15% of people
- It is associated with the buildup of Amyloid Beta in blood vessels and age-related cognitive decline during normal ageing
- Having one copy of E4 can increase your risk of AD by 200% while E2 actually reduces the risk by 40%
- A study with APOE knockout mice had significantly lower con. of HSV1 in the CNS than the wild-type mice, while the APOE4 mice had very high levels in the brain compared to the APOE3 animals.
Cytomegalovirus (CMV)

An introduction
What is CMV?

- Another member of the herpes family of viruses that affects everyone in two people.
- Several different studies to determine whether there is CMV in AD brains
- 1st Approach:
  - Used the PCR (Multiplex polymerase chain reaction) technique of DNA hybridisation
    - Found 36% in AD brains but 35% in controls.
    - BUT 93% found in vascular dementia patients
- 2nd Approach:
  - Examined serum antibody titres to the virus
    - Viral burden is associated with cognitive decline in elderly with vascular disease
What does it do?

- Deregulates inflammatory responses causes prolonged HSV1 reactivation
- Increases local inflammatory responses to various antigens, like Aβ
- Influences immune responses to other pathogens, triggering immune dysregulation involved in age related diseases
- **CMV infection facilitates the development of HSV1-associated AD, possibly via its effects on the immune system.**
HSV1 v. CMV Seropositivity

- HSV1 Seropositivity:
  - In children = impaired cognition
  - Middle aged = impaired reading and visuospatial processing
  - Elderly = immediate memory impairment

- CMV Seropositivity:
  - Only showed slower coding speed and impaired learning in middle age

This means that HSV1 has life course effects while CMV effects are restricted just to the middle aged.
1) Evidence of HSV1 reactivation in the human CNS

2) Effects of HSV1 Infection of mouse brains relating to AD, including evidence of reactivation
Evidence of HSV1 reactivation in the human CNS.
Evidence of HSV1 reactivation in the human CNS

Disclaimer:
- Difficult to study HSV1's role in AD due to the lack of current methods in detecting reactivation.
  - Specifically, if it occurs within a limited time frame and in very localized regions of the CNS.
- Detecting damage caused during HSE is easier than trying to study the reactivation of HSV1.
  - The Detection of HSE is done by seeking out HSV1 DNA in the CSF.

Study:
- 3200 CSF specimens from all ages submitted for HSV testing were randomly selected for analysis.

Results:

- HSV1 detected in 26 samples:
  - 9 male
  - 17 female
- HSV2 detected in 36 samples:
  - 13 male
  - 23 female

Conclusion:
- Overall, data showed twice as many HSV1 and HSV2 infections detected in females than males:
  - 22 male, 40 female

Correlation:
- Two-thirds of people with AD are women.
Evidence of HSV1 reactivation in the human CNS

Study:
- Pre-PCR study on HSV1 in post mortem human brain strongly suggested that HSV1 reactivates in the brain under conditions of immunosuppression.
  - Shown by patients with acute leukemia, who were immunosuppressed for part of their treatment.

Results:
- Immunosuppressed patients indicated the presence of HSV1 DNA in frontal and temporal cortices, but not in those who had not been immunosuppressed.

Correlation:
- Parts of the brain mostly affected in AD patients are the Frontal and temporal cortices.
Effects of HSV1 Infection of mouse brains relating to AD, including evidence of reactivation
Effects of HSV1 Infection of mouse brains relating to AD, including evidence of reactivation

Study:
- Examined CNS and trigeminal ganglia of HSV1 infected mice
  - Looking for several markers of inflammation and neurodegeneration to find evidence of asymptomatic reactivation of HSV1

Results:
- Detected viral ICP4 protein during acute, symptomatic phase of infection but also at 60 days post-infection.
  - 60 days post-infection is well after the start of the asymptomatic latent phase. **Indicating reactivation was occurring.**
  - **Note:** ICP4 stands for Infected-cell protein. ICP4 protein is the major transcriptional regulatory protein of HSV1.
Effects of HSV1 Infection of mouse brains relating to AD, including evidence of reactivation

Results (continued from previous slide):

- Simultaneous up-regulation of Interferon (INF) α and β and toll-like receptors (TLR’s)
  - This indicates neuroinflammatory action
    - Note: Interferons (INFs) are large class of proteins known as cytokines. Typically, a virus-infected cell will release (INFs) causing other cells to heighten their antiviral defenses.

- Also, there was an up-regulation of phospho-tau and caspase-3-cleaved tau.
  - These are indications of the early neurodegenerative processes.

Conclusion:

- Infection and reactavion causes up-regulation of INFs and TLRs indicating neuroinflammatory action. As well as the up regulation of phospho-tau and caspase-3-cleaved tau indicating early neurodegenerative processes.
Effects of HSV1 Infection of mouse brains relating to AD, including evidence of reactivation

Disclaimer:
- This study is on hepatitis, not HSV.

Study:
- Induced inflammation in mouse brain by viral infection (via endotoxin), to find if the inflammation modulated the AD-like features that develop in aged triple-transgenic (3xTg-AD) mice.

Procedure:
- A single dose of mouse hepatitis virus was used, thus creating an acute infection and a strong neuroinflammatory response within mouse brain.
Effects of HSV1 Infection of mouse brains relating to AD, including evidence of reactivation

Results:
- The findings of this mouse study between 3xTg-AD mice and non-Tg mice:
  - Immune responses did not differ between 3xTg-AD and non-Tg
  - Mortality did not differ between 3xTg-AD and non-Tg
  - Increased tau phosphorylation in the 3xTg-AD mice and not the non-Tg mice.
    - It also increased at 2 and 4 weeks past-injection, after the viral infection had been cleared, and when no short-term immune responses remained.

Conclusion:
- The evidence from this study suggests that similar effects of infection might occur in human brain. These infections could promote the development of AD by exacerbating pre-existing neuroinflammation and thereby exacerbating tau pathological features, hence accelerating cognitive decline.
Alzheimer's cell culture
Alzheimer's Hallmarks

- AD
  - Associated with neural loss and synaptic dysfunction
- AD cell culture
  - amyloid-β peptides, amyloid-protein precursor (APP) and hyperphosphorylated Tau protein
Alzheimer's Hallmarks

- Amyloid-β
- Amyloid-protein precursor (APP)
Studies on HSV1 infected cell cultures
Alzheimer's Hallmarks

- HSV-1
  - Accumulation of Aβ
  - Multiple types of cleavage APP
intracellular Ca$^{2+}$ signals
(from L-type Ca$^{2+}$ channels and InsP$_3$ receptors)
Epidemiological Studies
Epidemiological Studies

Background: Several studies have sought anti HSV IgM as well as IgG in serum from AD patients on the basis that the presence of IgM is associated with recent reactivation of HSV1, in contrast to IgG, which shows only that the person has been infected with HSV1. Serum antibody levels reflect the PNS, but it is unclear whether it or not it reflects the response from the CNS.

Study: Conducted by Letenneur et al. in 2008

- Revealed that elderly subjects who were IgM-positive were more likely to develop dementia within the following 14 years than those who were IgG-positive but not IgM-positive.

Result:

- Supports the concept that reactivation of the virus leads eventually to dementia.
Study Results: Cumulative Alzheimer’s Disease Rate According to Anti-HSV IgG or Anti-HSV IgM status
Epidemiological Studies

Study: Conducted by Feart et al. in 2011

- Found that high IgM levels are associated with low plasma levels of Aβ 1-40 and 1-42 - which are considered to be biomarkers of the disease, in that their decrease might reflect accumulation of Aβ in brain cells (and hence a reduction in plasma levels).

- Authors considered that virus reactivation might occur as a consequence of initial accumulation rather than being the cause of an accumulation of Aβ.

Results:

- Feart et al. concluded that their data supported the involvement of virus reactivations leading to alterations in APP (β-amyloid precursor protein) processing and eventually to Alzheimer’s Disease.

Important Note:

- No association of APOE-ε4 with IgM positivity was found in either of these studies which the authors attribute to the small number of people who were APOE-ε4 carriers and IgM positive.
Epidemiological Studies

Study: Conducted by Lovheim et al. in 2014

- They carried out a longitudinal prospective cohort study to investigate the possible relationship between HSV infection and Alzheimer’s Disease.
- They examined serum anti HSV antibodies and several neurophysiological, social and professional features in participants aged 25 to 95.
- The first cohort: One thousand participants were followed from 1990 to 1998 and then every 5 years thereafter.
- Other cohorts: Each comprising of one-thousand people were recruited 5 years, 10 years, and 15 years later.
- At all time points, numerous tests were used to assess each participant’s memory systems, problem-solving and decision making ability, and their health was examined in detail.

Results:

- The results were consistent with the two previous epidemiological studies revealing that the presence of anti HSV IgM antibodies, which indicate simplex virus reactivation, almost doubled the risk for Alzheimer’s.
- Presence of IgG antibodies did not indicate risk for Alzheimer’s.
Epidemiological Studies

Study: Conducted by Manuco et al. in 2014

- Examined serum anti-HSV 1 IgG titres of patients with mild Alzheimers, and cortical gray matter volume.
- As expected, IgG levels were similar for patients and age-matched normal people. However, high antibody levels were significantly more frequent in the patients, and correlated positively with cortical bilateral temporal and orbito-frontal gray matter volumes.
- Similar analyses were also carried out for Cytomegalovirus (CMV) antibodies

Results:

- No correlation was found between CMV antibodies and gray matter volumes thus strengthening the case for HSV-1 involvement.

Important Note:

- The authors commented that damage to the blood-brain barrier might enhance immune cell entry and suggested that HSV-1 specific antibodies play a protective role at early stages of AD by reducing HSV-1 activity in brain regions where the BBB is disrupted.
Genetic Studies
Several genome wide association (GWAS) has showed that HSV 1,2 associated with AD
- Virus enter to the neurons and regulate of defense mechanisms against the virus like apoptosis
- Herpes family are among the most probable pathogen candidates
- HSV1 binds to many cell proteins, modulating their expression, including many encoded by genes for various neurological diseases like AD
- Although antiviral drugs and vaccinations might affect incidence of many diseases, host protein mimicry suggests that the impacts of treatments could be impacted by autoimmune features
CMV vs HSV1

Does CMV cause AD?
HSV 1 cause a chronic low grade infection and can cause inflammation in the genital tract. It is a neurotropic virus → infect the CNS.
Possible Antiviral Treatments

(Memantine)
What are Antiviral Treatments?

Main Goal: To reduce the reproduction of viruses within the body.

Types of Antiviral Treatments:
- Herpes
- Hepatitis
- Influenza

Common Antiviral Drugs:

**Anti-herpes:**
- Acyclovir
- Valacyclovir
- Brivudin
- Cidofovir
- Many more...

**Anti-flu:**
- Oseltamivir
- Amantadine
- Zanamivir

**Anti-hepatitis:**
- Adefovir
- Entecavir
- Lamivudine
- Many more...

Source: Antiviral Drugs for Viruses Other Than HIV
1. Acyclovir (ACV) and Valacyclovir (VCV)

**Main Purpose:** ACV is the most common antiviral that is used to reduce HSV-1 induced P-tau and Aβ production, as well as to interrupt HSV-1 DNA replication.

**ACV vs VCV:**
VCV is the pro-drug form of ACV. VCV is a very safe and effective way to administer ACV in such a way that is of greater oral bioavailability.
Interesting Research regarding ACV and VCV

1. **Wozniak et al., 2011**: It was found that ACV reduced Aβ and P-tau accumulation in HSV1 infected cell cultures.
   
   **Key Idea**: It was recommended that treatment should aim to reduce the accumulation of Aβ back to normal levels, and ACV helps in this process.

1. **Kumar et al., 2010**: In a study on rabbits, it was found that HSV1 DNA was found within their tears because of asymptomatic shedding.
   
   **Key Idea**: Points to the possibility of treating AD patients with antiviral substances such as VCV and monitoring the progress of antiviral treatments from the tears and saliva (by using biomarkers).

1. **Prasad et al. 2012**: Found that VCV alleviated cognitive impairments in schizophrenia patients.

2. **Strandberg et al., Katan et al., and Tarter et al.**: Showed that HSV1 is associated with decreased cognition and was inferred that it is responsible for cognitive decline in some AD patients.
2. Intravenous Immunoglobulin (IVIG)

**Main Purpose:** It has been reasoned that IVIG is beneficial to AD patients because of its antiviral properties. It can help neutralize extracellular viruses, and along with lymphocytes, help destroy cells infected with HSV1.

**Production of IVIG:**

*Immunoglobulin* is any class of proteins that are present in the human immune system and that function as antibodies. IVIG are purified immunoglobulin G (IgG) proteins that are created from pooled human plasma, and contain 95% unmodified IgG. Administered as an intramuscular injection.
Interesting Research regarding IVIG

1. *Wozniak and Itzhaki, 2013*: It was found that IVIG was effective at reducing Aβ and P-tau levels. It works together well with ACV

**Key Idea:** If it was paired with ACV, it would be a beneficial treatment for AD.
3. Memantine

Main Purpose: An NMDA receptor antagonist. Has strong antiviral activity that is used to treat AD.

Benefits to Memantine:
It is a prescription drug that is often used to treat dementia as it can improve memory and the ability to function on a daily basis.

Source: Memantine for the Treatment of Dementia: A Review on its Current and Future Applications
Interesting Research regarding Memantine

1. Rive et al., 2013: Found that memantine was an important factor in reducing social agitation and aggression. **Key Idea:** This has big positive implications for the institutionalization and caregiving of AD patients.

1. Brison et al., 2014: Found that memantine interfered with replication of the HSV1 virus. **Key Idea:** It was proposed that it should be used to treat certain neurological diseases of possible viral cause.
Cog 163 students be like....

I'LL NEVER REMEMBER ALL THAT

Conclusion
1. We investigated the presence of HSV1 in the elderly brain and the implications.

2. The relationship between CMV and AD.

3. HSV1 in the CNS and implications on neural function.


5. CMV vs HSV1, the showdown!

6. Possible antiviral treatments.
There is good evidence that suggests that HSV1 is a causal factor in Alzheimer's disease. We have seen evidence from genetic studies, epidemiological studies, and mouse brain studies that help support this hypothesis!

Through studying antiviral treatment, it can be concluded that it is an effective and safe way to treat AD induced from a virus like HSV1.

Obviously, this leads to an improvement of life for AD patients!
Questions?
Thank you!