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HSV-1 Infection Is Associated with Reduced Alzheimer’s Disease Related Atrophy and Altered Brain Transcriptomic Signatures

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BACKGROUND & HYPOTHESIS

Early mechanistic reviews and multiscale post-mortem studies have linked higher Herpes simplex virus type 1 (HSV-1) abundance in brain tissue to disrupted molecular and clinical networks in Alzheimer’s disease (AD) , reinforcing the long-standing “viral risk” hypothesis (Itzhaki, 2017; Readhead et al., 2018). Population evidence, however, is beginning to shift this narrative. HSV-1 DNA is more **prevalent** in controls (Tejada M. et al., 2026). Recent antiviral trial showed that Valacyclovir showed **no efficacy** for the primary cognitive outcome (Devanand et al., 2026). In this cross-sectional analysis, we evaluated the association between HSV-1 presence and AD-related neuroanatomical changes using magnetic resonance imaging (MRI) data from 2956 participants of the Alzheimer’s Disease Sequencing Project (ADSP), including 967 AD cases, and 1989 cognitively normal controls. We also evaluated the association of HSV-1 DNA presence with microglial subtype composition and gene expression changes in DLPFC snRNA-seq data from 241 ROSMAP participants, including 124 AD cases and 117 controls.

Study question
Is HSV-1 DNA presence associated with AD-related neuroanatomical changes and altered microglial transcriptional programs, and does the association differ by AD status?

METHODS OVERVIEW

1 **HSV-1 DNA Detection:** (Tejada M., et al, 2024)

- WES/WGS reads aligned to the human reference genome (GRCh38); unmapped reads were retained
- Unmapped reads mapped against a curated viral genome database, including HSV-1
- HSV-1 aligned read counts normalized by each sample’s total sequencing depth
- Samples with ≥ 1 normalized HSV-1 read flagged as HSV-1 positive

2 **P-Value Adjustment:**

- To account for correlation among multiple association tests, we applied the P_{ACT} method of Conneely and Boehnke (2007).
- Under the global null, the vector of test statistics is modelled as multivariate normal with covariance matrix Σ estimated from the observed correlations.
- The adjusted p -value P_{ACT} is computed as the probability that at least one test in this MVN distribution yields a p -value as small as the observed minimum.

3 **Propensity Score Weighting:** (Rosenbaum P. R. & Rubin D. B., 1983)

- Estimate each subject’s propensity for HSV-1 exposure by fitting a logistic regression on observed covariates.
- Perform 1:1 nearest-neighbour matching without replacement.
- Evaluate post-match covariate balance via standardized mean differences (target < 0.1).
- Compute the average treatment effect by comparing MRI outcomes between matched HSV-1 positive and negative subjects.

4 **Cell Composition & Gene Expression Calculation:**

$$Y_i \sim \text{Binomial}(n_i, p_i),$$
$$\log\left(\frac{p_i}{1-p_i}\right) = \beta_0 + \beta_1 \text{HSV1}_i + \beta_2 \text{AD}_i + \beta_3 \text{HSV1}_i \text{AD}_i + \beta_4 \text{Age}_i + \beta_5 \text{Sex}_i$$

$$\text{Gene Expression}_{i,s}^k = \beta_0 + \beta_1 \text{HSV1}_i + \beta_2 \text{AD}_i + \beta_3 \text{Sex}_i + \beta_4 \text{Age}_i + \varepsilon_{i,s}$$

$$X_i = (\text{Age}_i, \text{Sex}_i, \text{ICV}_i, \text{ScannerBatch}_i)^\top$$
$$Y_i = \alpha + \tau D_i + \beta A_i + X_i^\top \delta + \epsilon_i$$
$$Y_i = \alpha + \tau D_i + \beta A_i + \gamma D_i A_i + X_i^\top \delta + \epsilon_i$$
$$Y_i = \alpha_s + \tau_s D_i + X_i^\top \delta_s + \epsilon_i, \quad s \in \{\text{Control}, \text{AD}\}$$

- Y_i denotes the MRI outcome for participant i , D_i indicates HSV-1 status, and A_i indicates AD diagnostic status.
- Scanner batches contain information on cohort names, MRI machine brands, and batch identifiers.

HSV-1 is linked to attenuated AD-related atrophy in selected brain regions.

Figure I. Joint effects of HSV-1 on MRI-derived brain measures. The left panel shows the HSV-1 effect in cognitively healthy controls, while the right panel shows the combined HSV-1 effect in AD patients. Red intervals indicate nominal significance ($p < 0.05$), and blue intervals indicate multiple-comparison significance ($p < 1.3 \times 10^{-3}$).

HSV-1 alters DLPFC microglial composition, with effects that vary by AD status.

Figure II. HSV-1 associated changes in microglial composition. HSV-1 effects on microglial subtype abundance, shown as log-odds estimates. Yellow indicates the pooled HSV-1 effect, pink indicates the effect in non-AD individuals, and blue indicates the effect in AD cases.

Inflammatory microglia show HSV-1 associated differential expression

Figure III. Volcano plots of HSV-1 associated differential gene expression in MG10 inflammatory microglia, stratified by AD status. Significant genes are colored by direction of effect, with genes significant in both groups shown in blue

Inflammatory microglia show HSV-1 associated immune pathway enrichment.

Figure IV. Gene Ontology gene set enrichment analysis for MG10 inflammatory microglia. Circles represent controls and triangles represent cases. Color indicates $-\log_{10}$ adjusted p -value

CONCLUSIONS

- HSV-1 presence is associated with **attenuated atrophy** on rostral middle frontal cortex
- HSV-1 presence **is not associated** with neurodegeneration.
- HSV-1 DNA presence **increases** lipid processing and phagocytic microglia in cases but decreases in controls.
- HSV-1 presence alters the microglial composition in DLPFC, and its effect changes with AD
- HSV-1 enhances inflammatory pathways in controls but not in AD cases.

HSV-1 DNA is associated with structural brain differences and immune related transcriptional signatures, with effects that may differ by AD.

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