**Virologica Sinica**

**Supplementary Data**

**Persistent inflammation and neuronal loss in the mouse brain induced by a modified form of attenuated herpes simplex virus type I**

Erlin Wang a, b, 1, Xinwei Huang a, 1, Yunshuang Ye b, 1, Shiqing Zou b, c, Guijun Chen b, Liping Yang b,

Nigel W. Fraser d, Fukai Bao e, Jumin Zhou b, f, \*, Xia Cao a, \*

a Key Laboratory of Second Affiliated Hospital of Kunming Medical University, Kunming, 650000, China

b Key Laboratory of Animal Models and Human Disease Mechanism of the Chinese Academy of Science/Key Laboratory of Healthy Aging Research of Yunnan Province, Kunming Institute of Zoology, the Chinese Academy of Sciences, Kunming, 650223, China

c Kunming College of Life Science, University of Chinese Academy of Sciences

d Department of Microbiology, Perelman School of Medicine, University of Pennsylvania, Philadelphia, PA19104, USA

e Department of Biology and Immunology, Kunming Medical University, Kunming, 650101, China

f KIZ/CUHK Joint Laboratory of Bioresources and Molecular Research in Common Diseases, Kunming, 650223, China.

1 Erlin Wang, Xinwei Huang, and Yunshuang Ye contributed equally to this work

\* Corresponding Author:

E-mail address: zhoujm@mail.kiz.ac.cn (J. Zhou); caoxia@kmmu.edu.cn (X. Cao).

ORCID: 0000-0003-1389-1087 (J.Zhou); 0000-0002-0453-9615 (X.Cao).

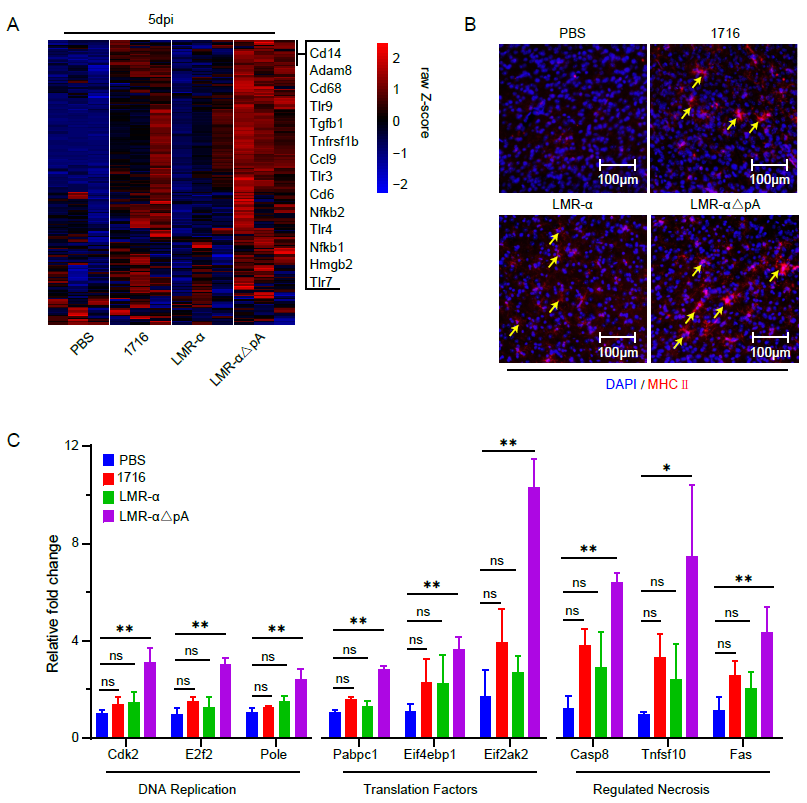


Figure S1. LMR-αΔpA infection activated greater levels of immune response. **A** Heatmap showing changes in the expression of immune response-associated genes in HSV-1 infected mice brains. Adjust *P*-value ≤ 0.05 and absolute log2 fold change ≥ 2 were used as thresholds. **B** MHC II (biolegend, clone M5/114.15.2) staining showed the activation of antigen presenting cells in 5dpi mouse brain. The yellow arrow indicates the positive staining signal. **C** RT-qPCR verified the transcription changes of selected genes in Fig. 2F. Data are displayed as mean ± sd. (\*\**P* < 0.01 using two-tailed *t*-tests).

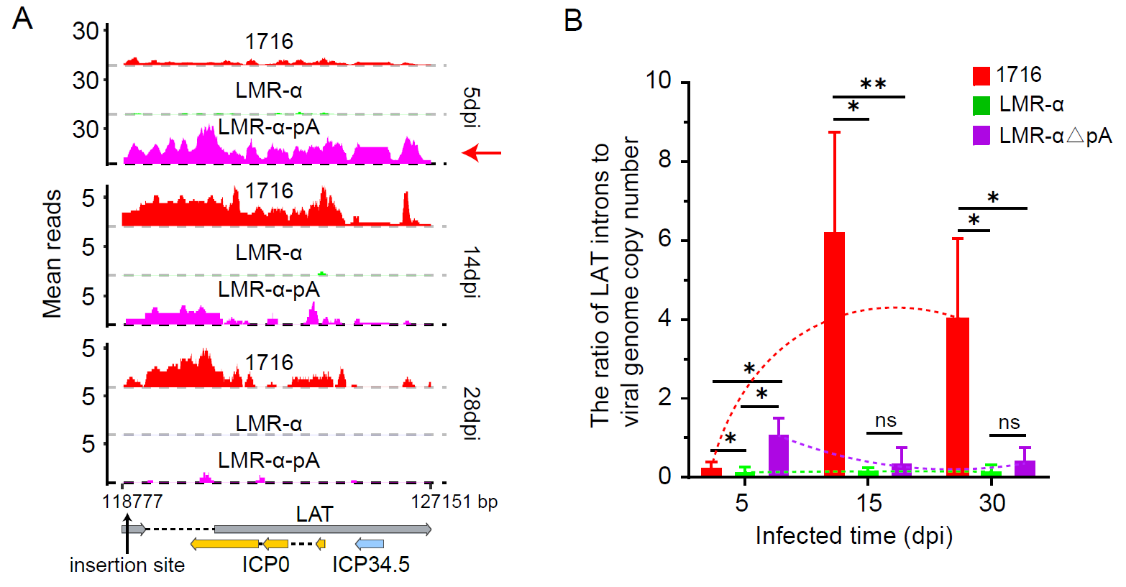


Figure S2. LMR-αΔpA has higher transcriptional activity in LAT region at 5 days post infection. **A** Coverage profiles of LAT region from each infection group. The arrows with red border were annotated the significant transcriptional differences. **B** LAT intron vs viral genome copies ratio among the three viruses across all time points. The dotted line shows a linear data fitting.

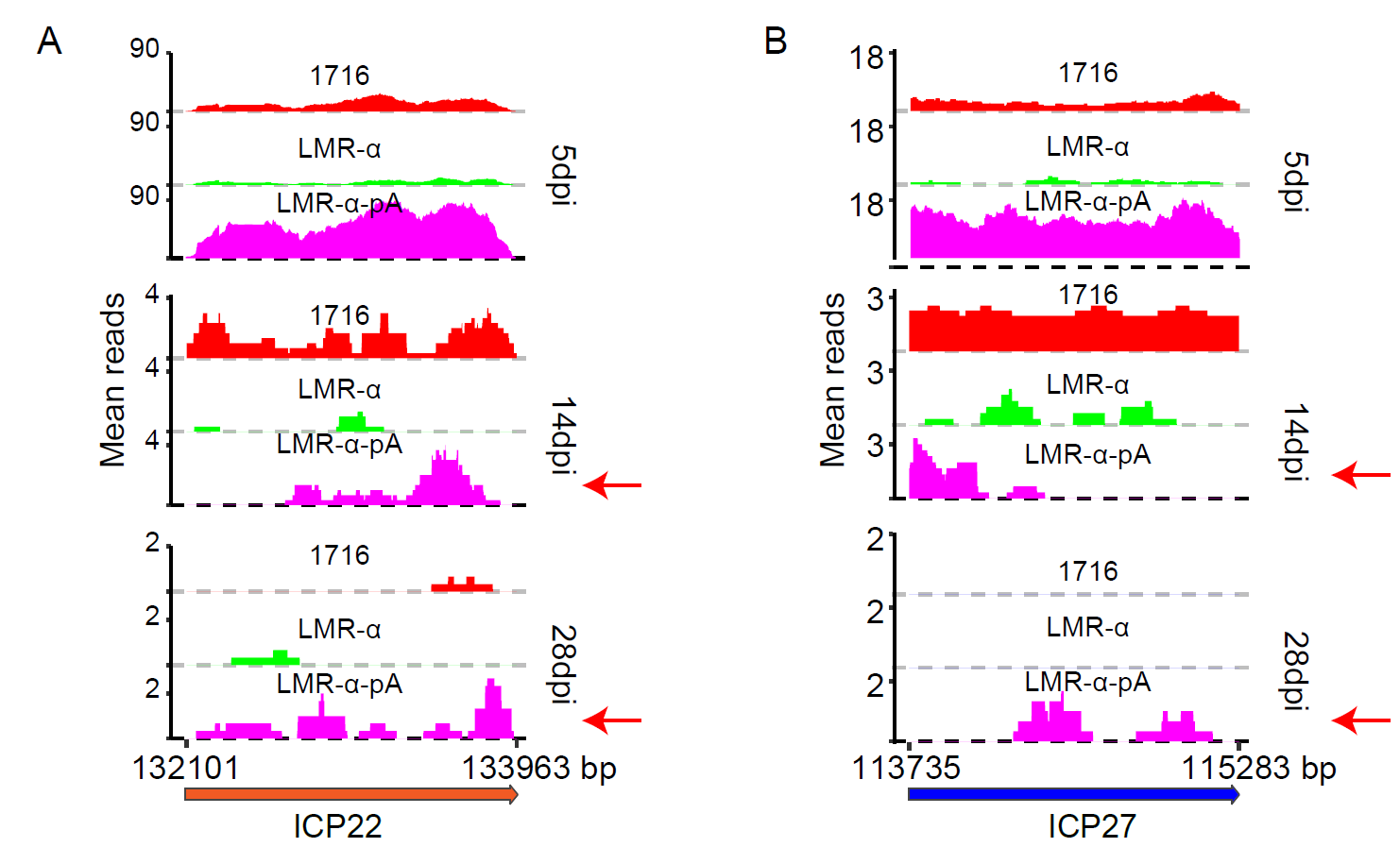


Figure S3. LMR-αΔpA infection had relatively stable transcriptional activities of *ICP22* and *ICP27* genes. Coverage profiles of ICP22 (**A**) and ICP27 **(B**) region from each infection group. The arrows with red border were annotated the significant transcriptional differences.

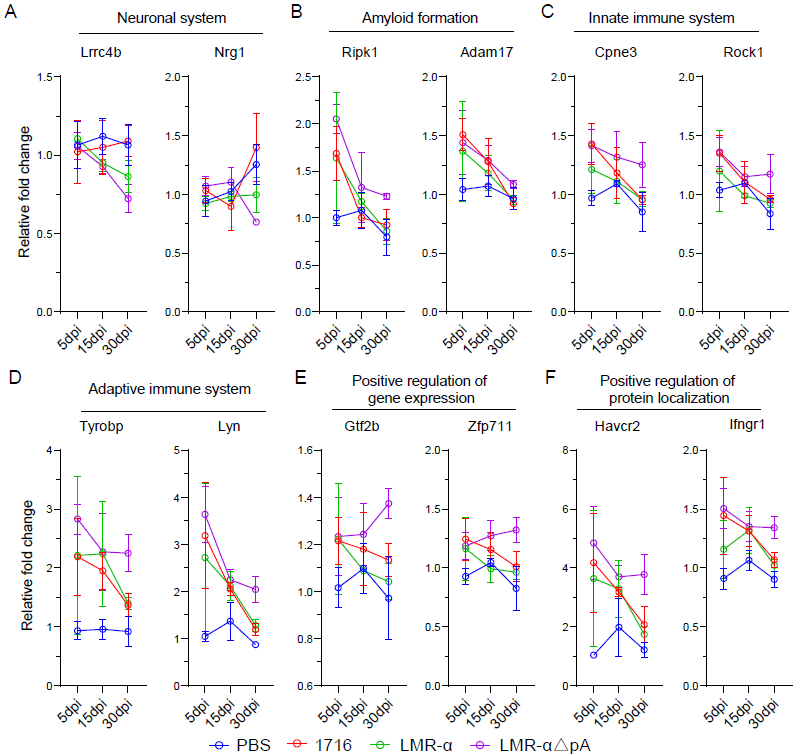


Figure S4. LMR-αΔpA infection inhibited nervous system related genes and led to continuous activation of immune genes. RT-qPCR verified the transcription changes of selected genes in Fig. 6C. GO: BP terms were labeled at the top of each panel.

Table S1. List of PCR primers.

|  |  |  |
| --- | --- | --- |
| **Target** | | **Primer sequence** |
| LAT intron | Fwd: | CCCACGTACTCCAAGAAGGC |
| Rev: | AGACCCAAGCATAGAGAGCCAG |
| Probe | CCCACCCCGCCTGTGTTTTTGTG |
| ICP27 | Fwd: | CGCCAAGAAAATTTCATCGAG |
| Rev: | ACATCTTGCACCACGCCAG |
| Probe: | CTGGCCTCCGCCGACGAGACG |
| Cdk2 | Fwd: | ATGGAGAACTTCCAAAAGGTGG |
| Rev: | CAGTCTCAGTGTCGAGCCG |
| E2f2 | Fwd: | ACGGCGCAACCTACAAAGAG |
| Rev: | GTCTGCGTGTAAAGCGAAGTG |
| Pole | Fwd: | TCAGCACTCAAGCGTCTGG |
| Rev: | AAATACGGCATATAGGGCAAGG |
| Pabpc1 | Fwd: | CCTCCTTCAGGTTACTTCATGGC |
| Rev: | GGACTTGGTCTTAGTTGAGCAAT |
| Eif4ebp1 | Fwd: | GGGGACTACAGCACCACTC |
| Rev: | GTTCCGACACTCCATCAGAAAT |
| Eif2ak2 | Fwd: | ATGCACGGAGTAGCCATTACG |
| Rev: | TGACAATCCACCTTGTTTTCGT |
| Casp8 | Fwd: | TGCTTGGACTACATCCCACAC |
| Rev: | GTTGCAGTCTAGGAAGTTGACC |
| Tnfsf10 | Fwd: | ATGGTGATTTGCATAGTGCTCC |
| Rev: | GCAAGCAGGGTCTGTTCAAGA |
| Fas | Fwd: | GCGGGTTCGTGAAACTGATAA |
| Rev: | GCAAAATGGGCCTCCTTGATA |
| Lrrc4b | Fwd: | ATCCCTGTCAACACCCGCTA |
| Rev: | CCCACCTCGATCTTTCGCAC |
| Nrg1 | Fwd: | TTCCCATTCTGGCTTGTCTAGT |
| Rev: | CCAGGGTCAAGGTGGGTAG |
| Ripk1 | Fwd: | GACAGACCTAGACAGCGGAG |
| Rev: | CCAGTAGCTTCACCACTCGAC |
| Adam17 | Fwd: | ACCACTTTGGTGCCTTTCGT |
| Rev: | GTCGCAGACTGTAGATCCCTT |
| Cpne3 | Fwd: | GCCCAGTGTGTCACGAAGG |
| Rev: | AAGGGTCCGACTTTGATGTTAC |
| Rock1 | Fwd: | GACTGGGGACAGTTTTGAGAC |
| Rev: | ATCCAAATCATAAACCAGGGCAT |
| Tyrobp | Fwd: | CCCAAGATGCGACTGTTCTTC |
| Rev: | GTCCCTTGACCTCGGGAGA |
| Lyn | Fwd: | ATTGTGGTGGCCTTATACCCT |
| Rev: | ACCATTCCCCATGCTCTTCTA |
| Gtf2b | Fwd: | ACCAGCCGTTTGGATGCTC |
| Rev: | AGATCATATCGCCAGCTCTGTA |
| Zfp711 | Fwd: | AAGTCTTGGATTGCACACTTCA |
| Rev: | GGCCATGATCGAGAGTTATCCC |
| Havcr2 | Fwd: | TCAGGTCTTACCCTCAACTGTG |
| Rev: | GGCATTCTTACCAACCTCAAACA |
| Ifngr1 | Fwd: | GTGGAGCTTTGACGAGCACT |
| Rev: | ATTCCCAGCATACGACAGGGT |