**SUPPLEMENTARY MATERIALS**



**Fig.S1 RNA antisense purification against circSP100.**

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**Fig.S2 CircSP100 inhibits DNA-PK mediated innate immune response in HCMV infected HELF cells**

**Table S1. Primers used in RT-PCR, northern blot, RT-qPCR, and subcellular fractionation**

|  |  |  |
| --- | --- | --- |
| Primer  | Sequence (5'→3') | Product |
| **RT-PCR**  |
| circPLEKHM1-F | GCTTCAGGTAGCACTCCATCAGG | 208 bp |
| circPLEKHM1-R | GGCTCCAAGAGTCCAGACCATTG |
| circSP100-F1 | GCCTCCAACTAAGTCTTGAACAAGG | 713 bp |
| circSP100-R1 | CAGACCTCTCCTCCCTCTCTT |
| circTRIO-F | GGAATGTGACCCCGAAGAGAT | 209 bp |
| circTRIO-R | TTGTCAGGGAATCGGAGCTGCCG |
| circMAP3K1-F | GCTCCATCTCGTAACACCATCCAG | 182 bp |
| circMAP3K1-R | GCGGTTTGTTTCCTCAGGGCTAT |
| linear SP100-F1 | CCAGGAGGCTGAATGAATGT | 530 bp |
| linear SP100-R1 | CCAAGTCAGGCTTCGAAAAG |
| GAPDH-F1 | TGGTGAAGACGCCAGTGGA | 137 bp |
| GAPDH-R1 | GCACCGTCAAGGCTGAGAAC |
| **Northern bolt** |
| sense F | TAATACGACTCACTATAGGGGATTCGCTAGGAAGCCAACAA | 420 bp |
| sense R | CTTCTCCAGTTCACTAAGAACATTG |
| antisense F | GATTCGCTAGGAAGCCAACAA | 420 bp |
| antisense R | TAATACGACTCACTATAGGGCTTCTCCAGTTCACTAAGAACATTG |
| **RT-qPCR** |
| circSP100-F2 | GATGAAGAAAGATGTTCACGG | 120 bp |
| circSP100-R2 | GAAATGTCTTTTTTATTGCATTTG |
| linear SP100-F2 | AACAAGAACCCGTGGAGTTG | 320bp |
| linear SP100-R2 | ATGTCCTGCACAAACCCTTC |
| **Subcellular fractionation, RAP control** |
| GAPDH-F2 | GTCATCAATGGAAATCCCATCA | 320 bp |
| GAPDH-R2 | TGAGTCCTTCCACGATACC |
| circSP100-F3 | GATTCGCTAGGAAGCCAACAA | 380 bp |
| circSP100-R3 | CTTCTCCAGTTCACTAAGAACATTG |
| U2 snRNA-F | ATCTGATACGTCCTCTATCCGA | 108bp |
| U2 snRNA-R | CTGCAATACCAGGTCGATGC |

Primers listed above are related to Figure 3, Figure 4, and Figure S1.

**Table S2. Primers used for RNA probe preparation in RNA antisense purification assay.**

|  |  |  |
| --- | --- | --- |
| Primer  | Forward (5'→3') | Reverse (5'→3') |
| P1 | TGCAGGGGGAGTTTTATTTGATGTGCTCAAAAGGCTGA | AGACTGCCACGGTCATAAGCCGTGAACATCTTTCTTCATCA |
| P2 | TGCAGGGGGAGTTTTATTTGAAGGCTGAGCCAACAGA | AGACTGCCACGGTCATAAGCCCTGGTCTTCCGTGAACA |
| P3 | TGCAGGGGGAGTTTTATTTGCAACAGAGTCCTGCGAAC | AGACTGCCACGGTCATAAGCTCATCTACACCCTGGTCTTC |
| P4 | TGCAGGGGGAGTTTTATTTGCTGCGAACAAATTGCTGT | AGACTGCCACGGTCATAAGCGAGCAGCCTGTCATCTAC |
| P5 | TGCAGGGGGAGTTTTATTTGATTGCTGTCCAAGTGAATAATG | AGACTGCCACGGTCATAAGCCAATGTCATAGAGCAGCCT |
| P6 | TGCAGGGGGAGTTTTATTTGAAGTGAATAATGGGGATGCT | AGACTGCCACGGTCATAAGCTGCTTGAATACAATGTCATAGAG |
| P7 | TGCAGGGGGAGTTTTATTTGAATGGGGATGCTGGAAGG | AGACTGCCACGGTCATAAGCTCTTTTGAAGTGCTTGAATACAA |
| P8 | TGCAGGGGGAGTTTTATTTGGGAAGGGAGATGCCCTG | AGACTGCCACGGTCATAAGCCCACCTTATTTCTTTTGAAGTG |
| P9 | TGCAGGGGGAGTTTTATTTGAGATGCCCTGCCCGTT | AGACTGCCACGGTCATAAGCTTTGAAATCTCCACCTTATTTCTT |
| P10 | TGCAGGGGGAGTTTTATTTGGTTGCCCTGTGATGAAGAA | AGACTGCCACGGTCATAAGCTTTTATTGCATTTGAAATCTCCAC |
| P11 | TGCAGGGGGAGTTTTATTTG GATGAAGAAAGATGTTCACGG | AGACTGCCACGGTCATAAGCGAAATGTCTTTTTTATTGCATTTG |
| Tag | TGCAGGGGGAGTTTTATTTG | AGACTGCCACGGTCATAAGC |
| Tag+T7 | —————— | GGATTCTAATACGACTCACTATAGGGAGACTGCCACGGTCATAAGC |

Primers listed above are related to Figure 5 and Figure S1.

**Table S3. List of circRNAs detected by RNA-seq in mock-infected and HCMV-infected HELFs**

**Table S4. Detailed information about circSP100 binding proteins captured by RAP in un-crosslinked and UV-crosslinked lysis.**