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**Supplementary Data**

**FoxJ1 inhibits African swine fever virus replication and viral S273R protein decreases the expression of FoxJ1 to impair its antiviral effect**

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**Supplementary Table S1.** TheqPCR primers and siRNA sequences used in this study.

|  |  |  |
| --- | --- | --- |
| Primer | Sequence (5’–3’) | Target gene |
| pIFN-β-F | GCTAACAAGTGCATCCTCCAAA | Porcine *IFN-β* gene |
| pIFN-β-R | AGCACATCATAGCTCATGGAAAGA |  |
| pISG15-F | GATCGGTGTGCCTGCCTTC | Porcine *ISG15* gene |
| pISG15-R | CGTTGCTGCGACCCTTGT |  |
| pISG54-F | CTGGCAAAGAGCCCTAAGGA | Porcine *ISG54* gene |
| pISG54-R | CTCAGAGGGTCAATGGAATTCC |  |
| pISG56-F | TTAGAAAACAGGGTCTTGGAGGAG | Porcine *ISG56* gene |
| pISG56-R | CGTAAGGTAATACAGCCAGGCATA |  |
| pGAPDH-F | ACATGGCCTCCAAGGAGTAAGA | Porcine *GAPDH* gene |
| pGAPDH-R | GATCGAGTTGGGGCTGTGACT |  |
| pFoxJ1-FpFoxJ1-R | TACTCCTATGCCACGCTCATGCGGAAGTAGCAGAAGTTGTC | Porcine *FoxJ1* gene |
| mFoxJ1-FmFoxJ1-R | GTGGACTACGCCACCAATCCTAGATGGCCGACAGGGTGAT | Monkey *FoxJ1* gene |
| mGAPDH-FmGAPDH-R | CCTGCACCACCAACTGCTTACATGAGTCCTTCCACGATACCA | Monkey *GAPDH* gene |
| mISG15-FmISG15-R | TGGACAAATGCGACGAACCCCCGCTCACTTGCTGCTT | Monkey *ISG15* gene |
| mISG54-FmISG54-R | ACCGAACAATGCCTACCTGTGAGCCACAGCGTGTCCTA | Monkey *ISG54* gene |
| mISG56-FmISG56-R | TGTATTACCACATGGGCAGACATCCAGGCGATAGGCAGAG | Monkey *ISG56* gene |
| mIFN-β-FmIFN-β-R | GACATCCCTGAGGAGATTAAGATGTTCTGGAGCATCTCATAG | Monkey *IFN-β* gene |
| ASFV p72 (B646L)-F | TGCGATGATGATTACCTT | ASFV *B646L* gene |
| ASFV p72(B646L)-R | ATTCTCTTGCTCTGGATAC |  |
| ASFV p30(CP204L)-F | CTCCGATGAGGGCTCTTGCT | ASFV *CP204L* gene |
| ASFV p30(CP204L)-R | AGACGGAATCCTCAGCATCTTC |  |
| FoxJ1-siRNA-911(FoxJ1-siRNA)-FFoxJ1-siRNA-911(FoxJ1-siRNA)-R | CUCAAAGGCAACUUCGACUTTAGUCGAAGUUGCCUUUGAGTT | Porcine *FoxJ1* gene |
| FoxJ1-siRNA-911(FoxJ1-siRNA1)-FFoxJ1-siRNA-911(FoxJ1-siRNA1)-R | CUCAAAGGCAACUUCGACUTTAGUCGAAGUUGCCUUUGAGTT | Monkey *FoxJ1* gene |

**Supplementary Table S2.** The top ten genes that were most significantly down-regulated at each time point after ASFV infection.

|  |  |  |  |
| --- | --- | --- | --- |
| Gene category | 12 hpi | 24 hpi | 48 hpi |
| Gene | log2Fold Change | Gene | log2Fold Change | Gene | log2Fold Change |
| Downregulated | *LOC100522201* | -12.873  | *LOC100522201* | -12.992  | *LOC100522201* | -12.735  |
| *FOSB* | -9.033  | *FOSB* | -11.965  | *FOXJ1* | -12.464  |
| *FOXJ1* | -8.936  | *FOXJ1* | -10.863  | *FOSB* | -11.374  |
| *LOC100038328* | -8.756  | *RRAD* | -9.777  | *RRAD* | -10.521  |
| *FOS* | -8.446  | *ID1* | -9.281  | *LOC396781* | -9.973  |
| *EGR4* | -8.065  | *MT1D* | -9.072  | *CLEC12A* | -9.371  |
| *KIF18B* | -7.779  | *FAM83D* | -9.008  | *METTL24* | -9.138  |
| *PLIN1* | -7.772  | *ARC* | -8.940  | *LOC110258822* | -9.120  |
| *LOC102162420* | -7.567  | *KIF18B* | -8.861  | *LOC100038328* | -8.619  |
| *E2F8* | -7.523  | *THBD* | -8.775  | *PLIN1* | -8.596  |



Supplementary Figure S1. Screening of ASFV proteins that interacted with FoxJ1. iPAM cells were co-transfected with empty vector (2 µg/well) or Myc-FoxJ1 (2 µg/well) expressing plasmids and the indicated ASFV protein-expressing plasmids (2 µg/well) for 24 h. The expression of the indicated viral proteins and FoxJ1 was then detected by Western blotting.



Supplementary Figure S2. Working concentration of MG132, Z-VAD-FMK, CQ, and NH4Cl in iPAM cells. **A** The effects of inhibitors on the MGF-505-7R-mediated destabilization of JAK2. iPAM cells were transfected with MGF505-7R expressing plasmids or empty vector, and JAK2 expressing plasmids. MG132 (50 μmol/L) or NH4Cl (20 mmol/L) was added after transfection for 18 h. After 6 h incubation, the expression of HA-JAK2 was detected by Western blotting. **B** iPAM cells were transfected with MGF505-2R or E165R expressing plasmids, and vector or FoxJ1 expressing plasmids. MG132 (50 μmol/L) or NH4Cl (20 mmol/L) was added after18 h transfection. After 6 h treatment, the expression of Flag-MGF505-2R or Flag-E165R was detected by Western blotting. **C** iPAM cells were transfected with SVA-2B expressing plasmids or empty vector plasmids, and MAVS expressing plasmids. Z-VAD-FMK (50 μmol/L) and CQ (100 μmol/L) was added after 18 h transfection. After 6 h treatment, the expression of HA-MAVS was detected by Western blotting. **D** iPAM cells were transfected with MGF505-2R or E165R expressing plasmids, and vector or FoxJ1 expressing plasmids. Z-VAD-FMK (50 μmol/L) was added after 18 h transfection. After 6 h incubation, the expression of Flag-MGF505-2R or Flag-E165R was detected by Western blotting.