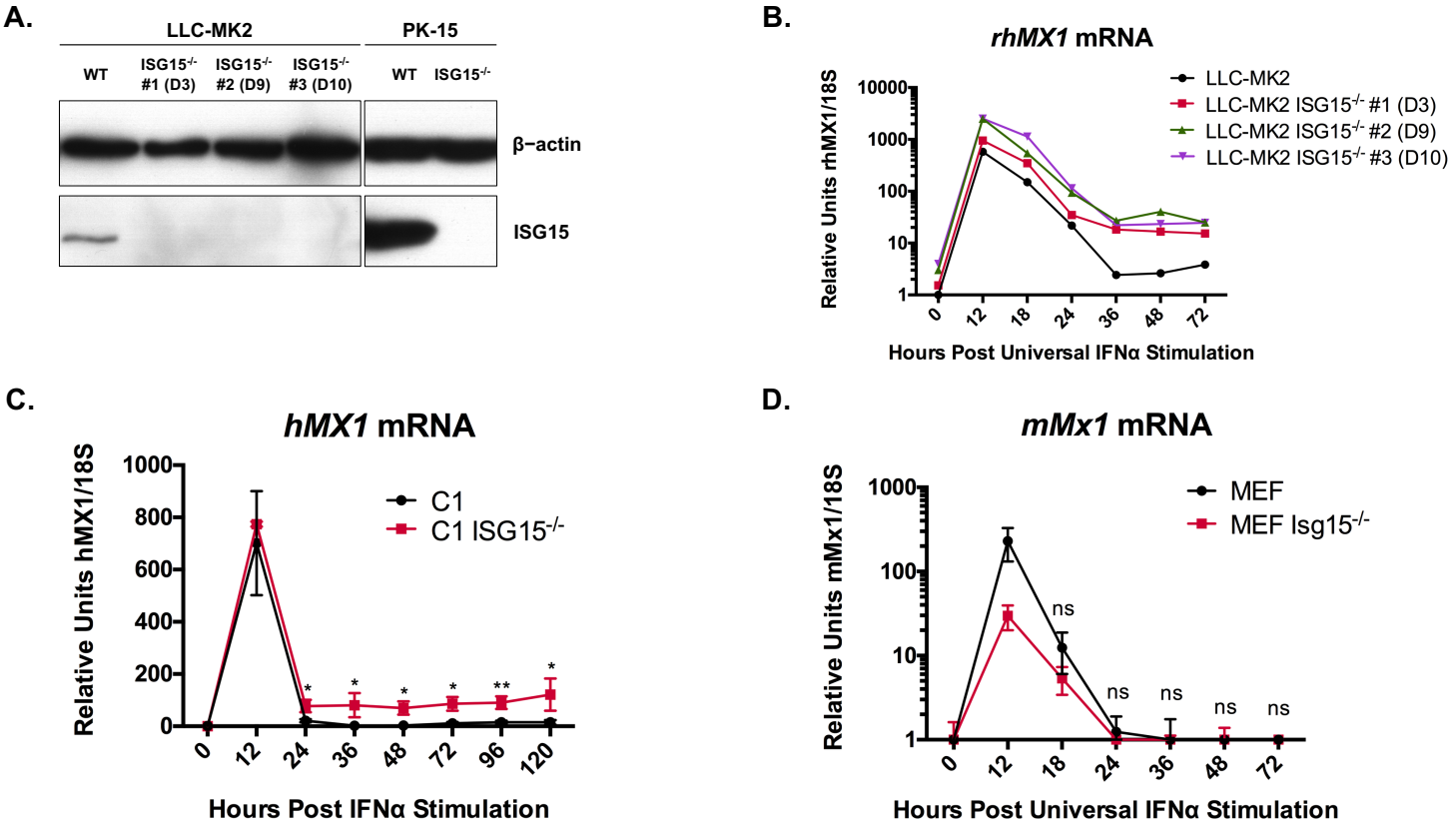

Supplementary Figure 4 | (A) Successful knockout of ISG15 in LLC-MK2 and PK-15 cells. WT and ISG15-KO LLC-MK2 and PK-15 cells were treated with 100 U/mL universal IFNα for 8 hours. Whole cell lysates were analyzed with western blotting using an ISG15 antibody with cross-species reactivity (ab233071). D3, D9, and D10 denote three different monoclonal cell populations of LLC-MK2 post CRISPR/Cas9-mediated knockout of ISG15. **(B-D) ISG expression of IFN-primed ISG15-deficient rhesus macaque, human, and murine cells.** WT and ISG15-knockout LLC-MK2 cells, hTert-immortalized human fibroblasts from a healthy donor (C1) and isogenic ISG15-KO C1 cells, and WT and ISG15-deficient MEFs were stimulated with 1000 IU/mL IFN-α2b or 100 U/mL universal IFN-α for 12 hours, washed with PBS, and allowed to rest for various lengths of time. Rhesus macaque *MX1* (*rhMX1*) **(B)**, human *MX1* (*hMX1*) **(C)**, and murine *Mx1* (*mMx1*) **(D)** mRNA levels relative to *18S* eukaryotic rRNA at indicated hours post stimulation were analyzed. Statistical comparison was performed with unpaired t tests. **B** shows a single experiment. **C** and **D** show a representative experiment with biological triplicates of two-three performed. Error bars, s.d. *p<0.05, **p<0.01, ***p<0.001, ****p<0.0001. NS, not significant.