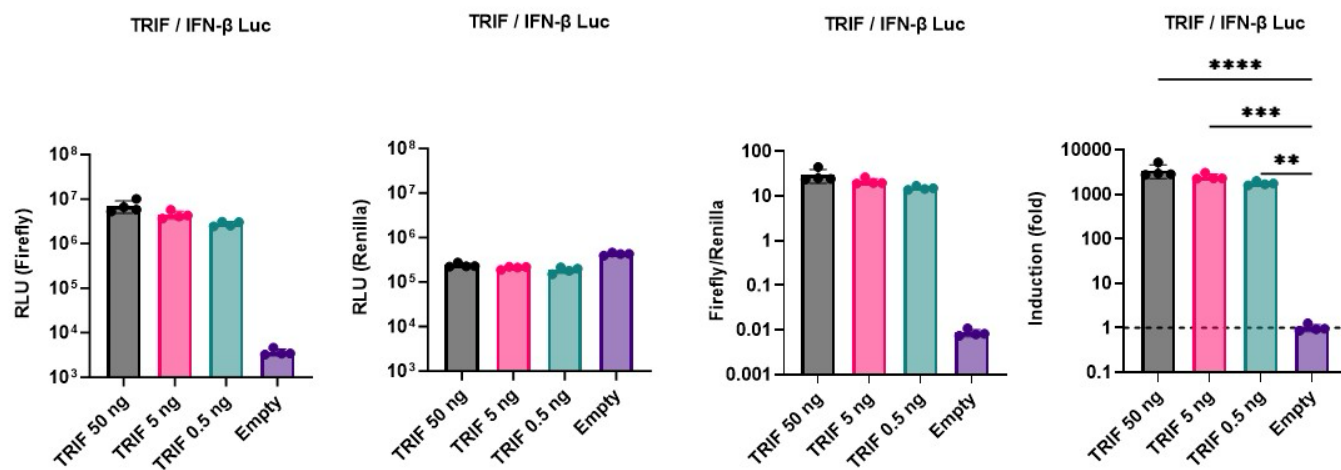
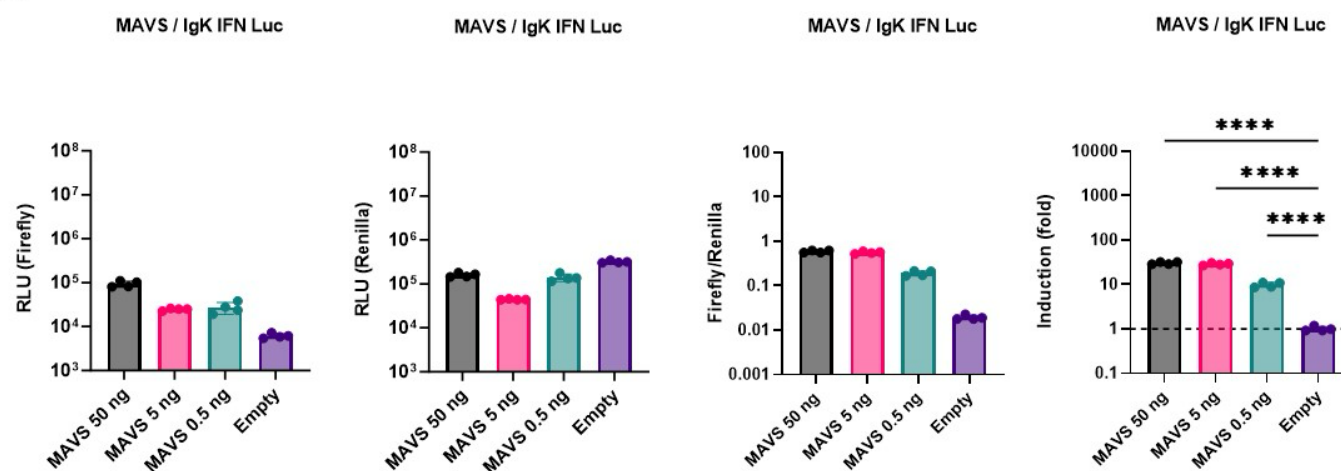




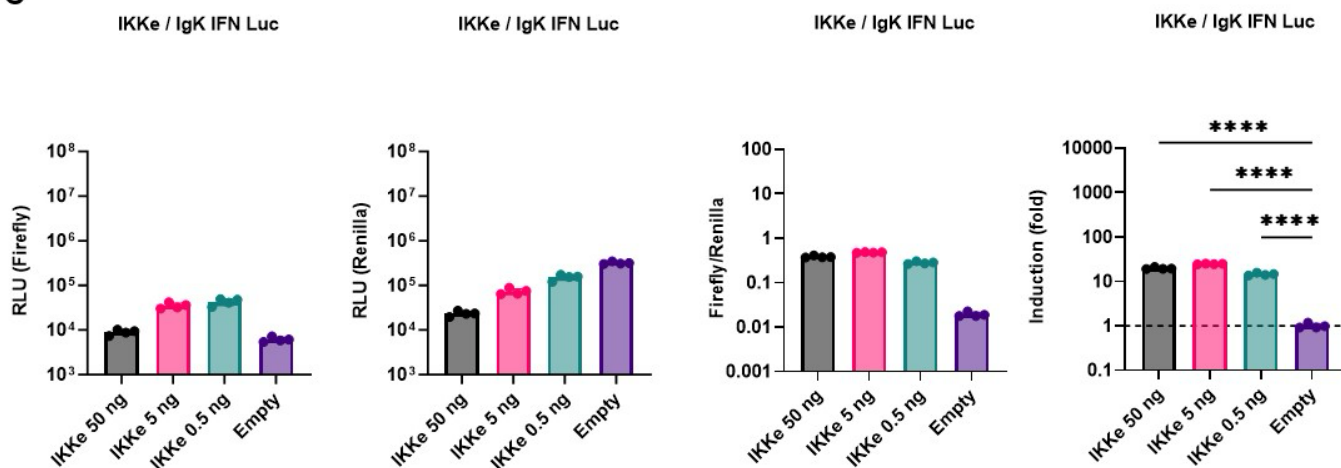
**A**

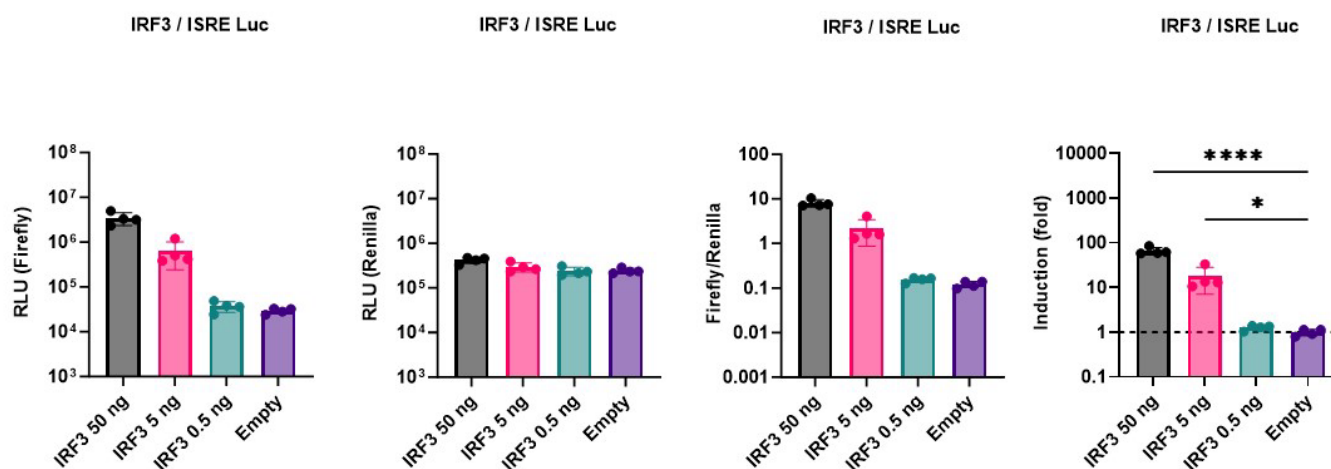
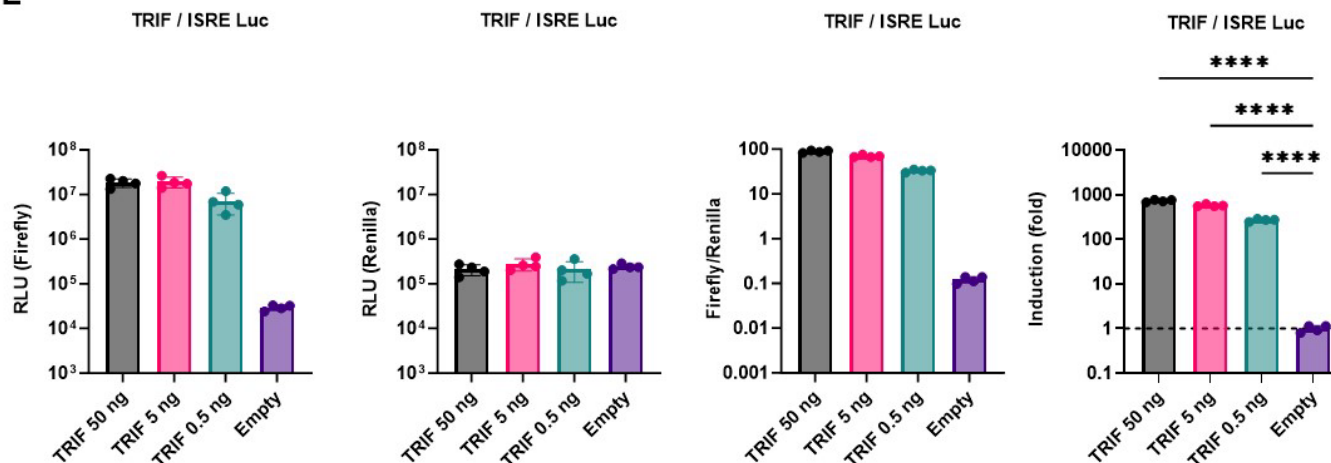


**B**



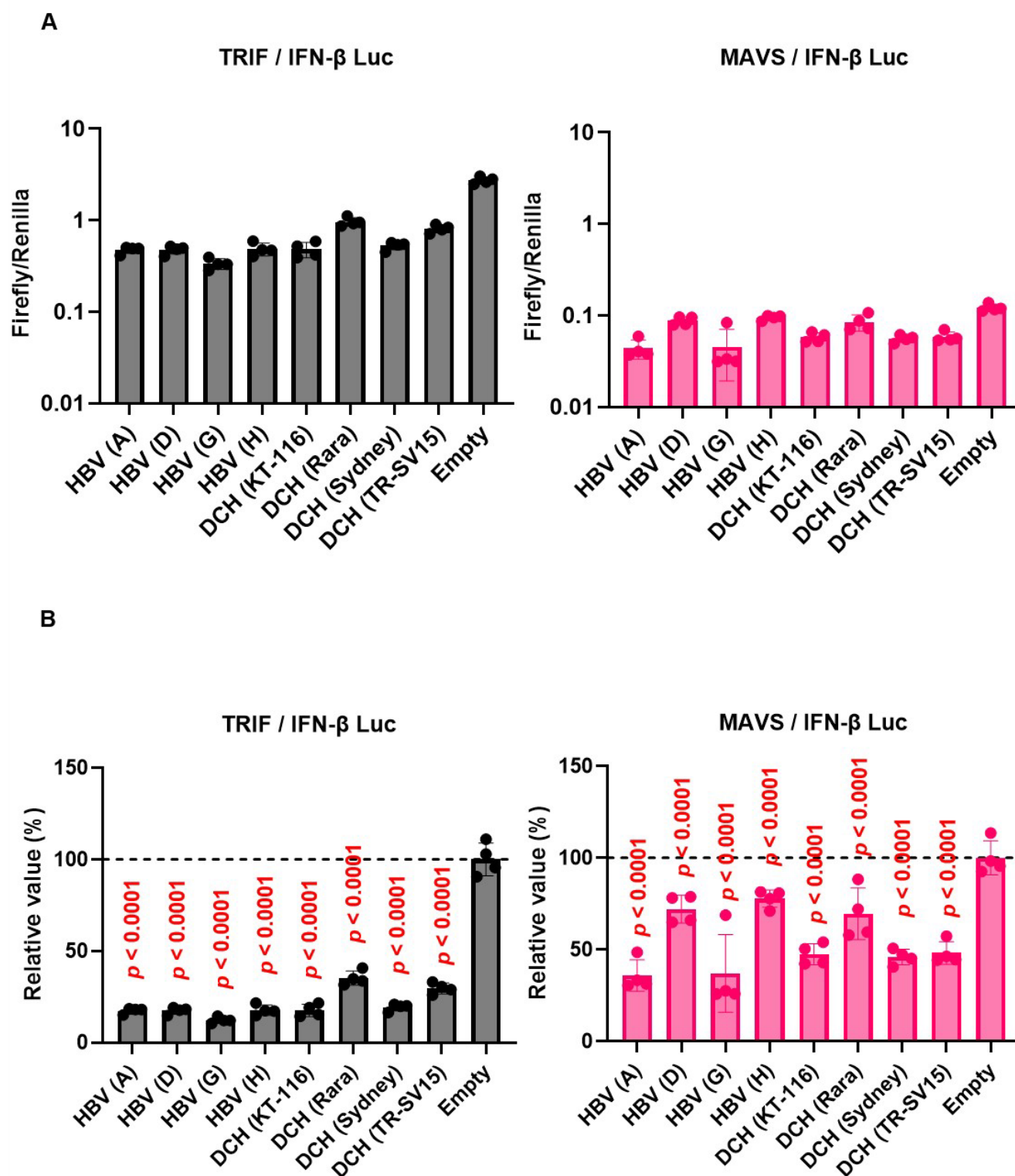
**C**



**D****E**

### Supplementary Figure S1. Induction of Firefly luciferase.

Co-transfection of the following plasmids (A) IFN- $\beta$  Luc and TRIF, (B) IgK-IFN Luc and MAVS, (C) IgK-IFN Luc and IKKe, (D) interferon-stimulated response element (ISRE) Luc and IRF3, and (E) ISRE Luc and TRIF. Differences between IFN-stimulating plasmid and empty plasmid were examined by one-way ANOVA followed by Dunnett's multiple comparison test. \*\*\*\* $p$  < 0.0001, \*\*\* $p$  < 0.001, and \* $p$  < 0.05.

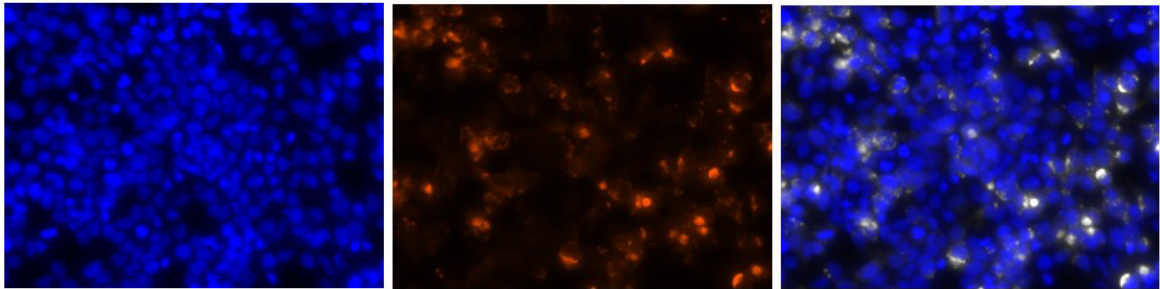


**Supplemental Figure S2. *Orthohepadnavirus* X protein inhibits IFN- $\beta$  induction mediated by TRIF and MAVS. (A)** Raw data of the luciferase reporter assay. The RLU of Firefly luciferase was divided by the RLU of Renilla luciferase. **(B)** Relative value of IFN- $\beta$  luciferase reporter assay. Differences between *Orthohepadnavirus* X proteins and “Empty” were examined by one-way ANOVA followed by Dunnett’s multiple comparison test.

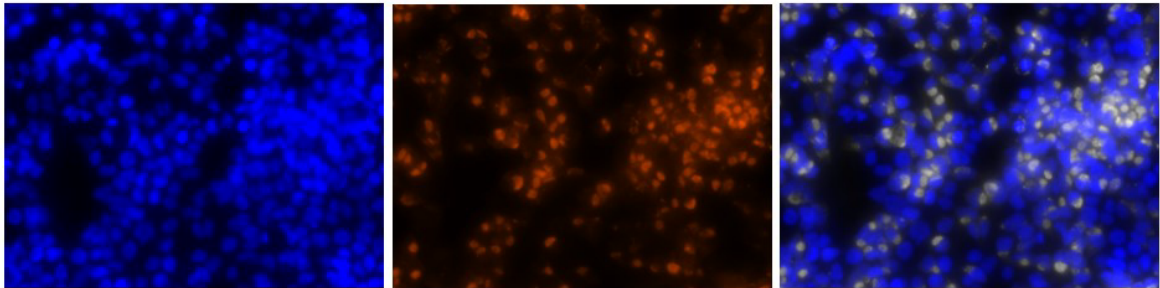
**A**

	<b>NucBlue</b>	<b>X protein (Anti-HA)</b>	<b>Merged</b>
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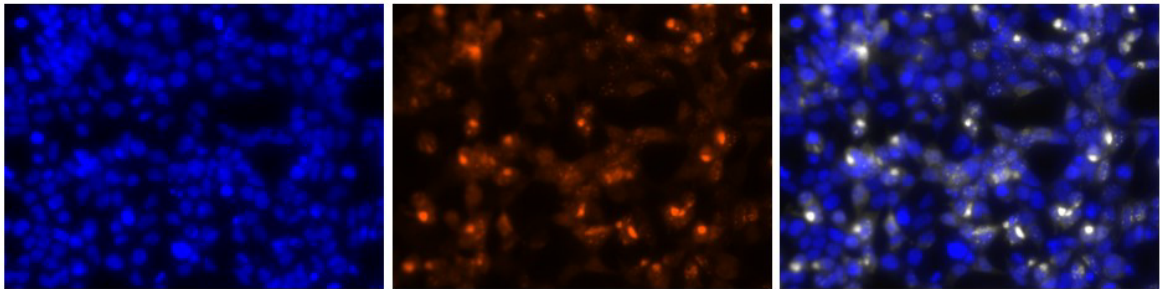
**HBV (A)**



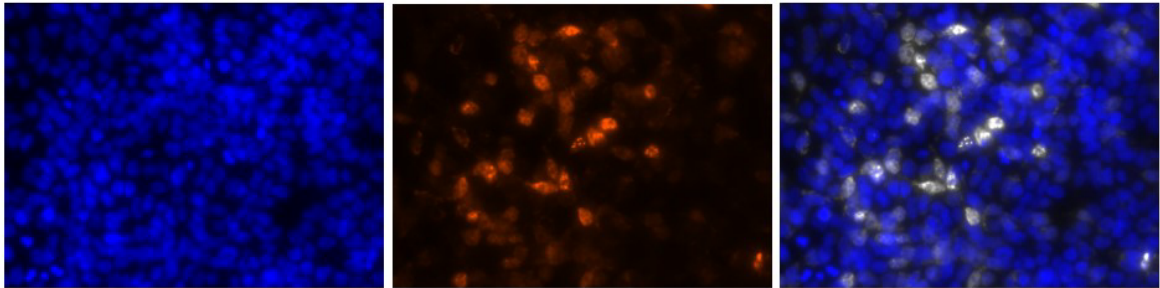
**DCH (KT-116)**



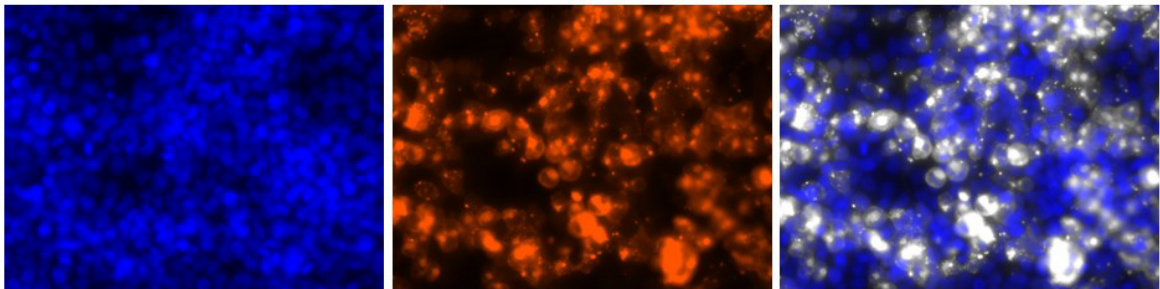
**DDHBV**



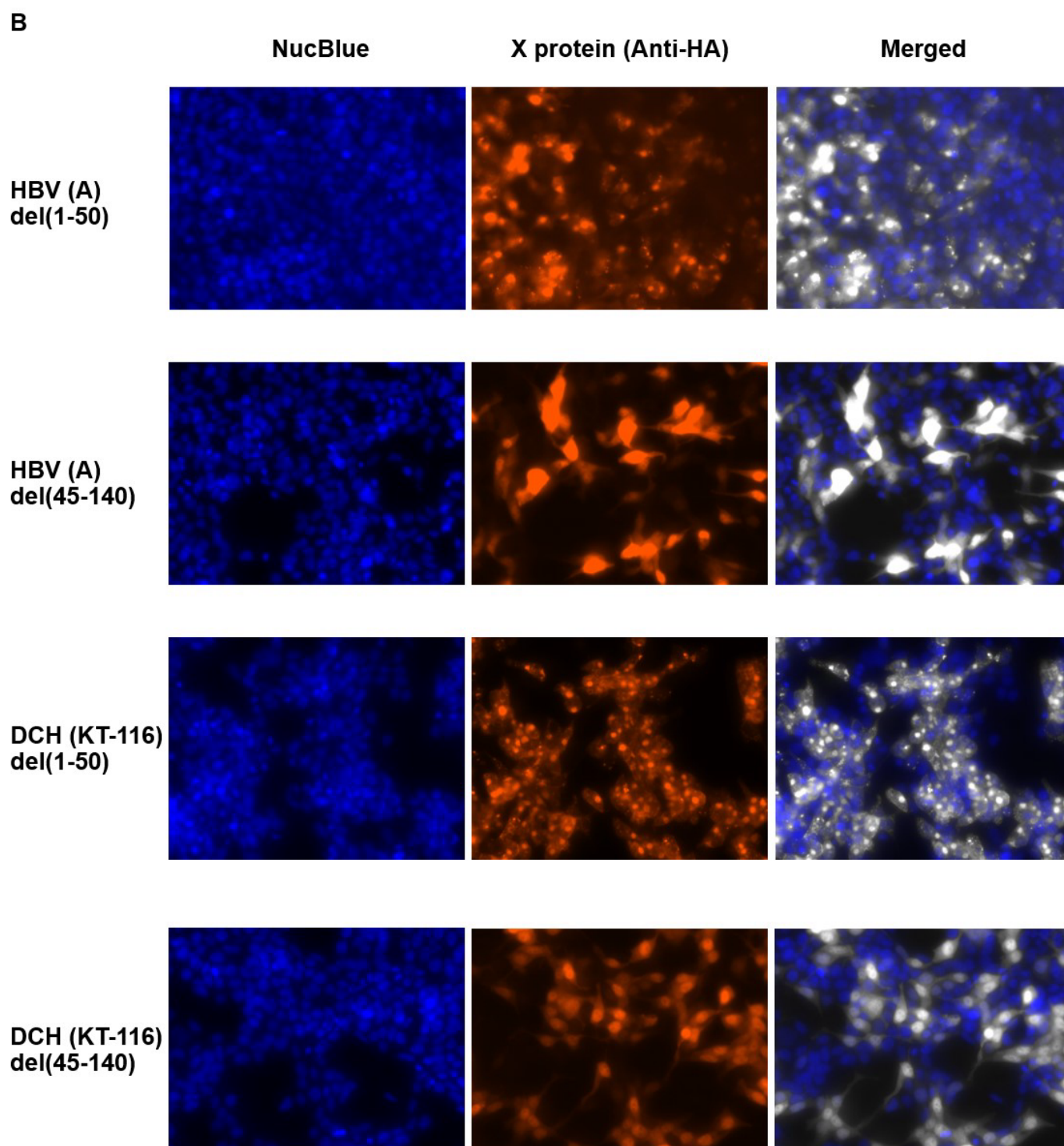
**AGSHBV**



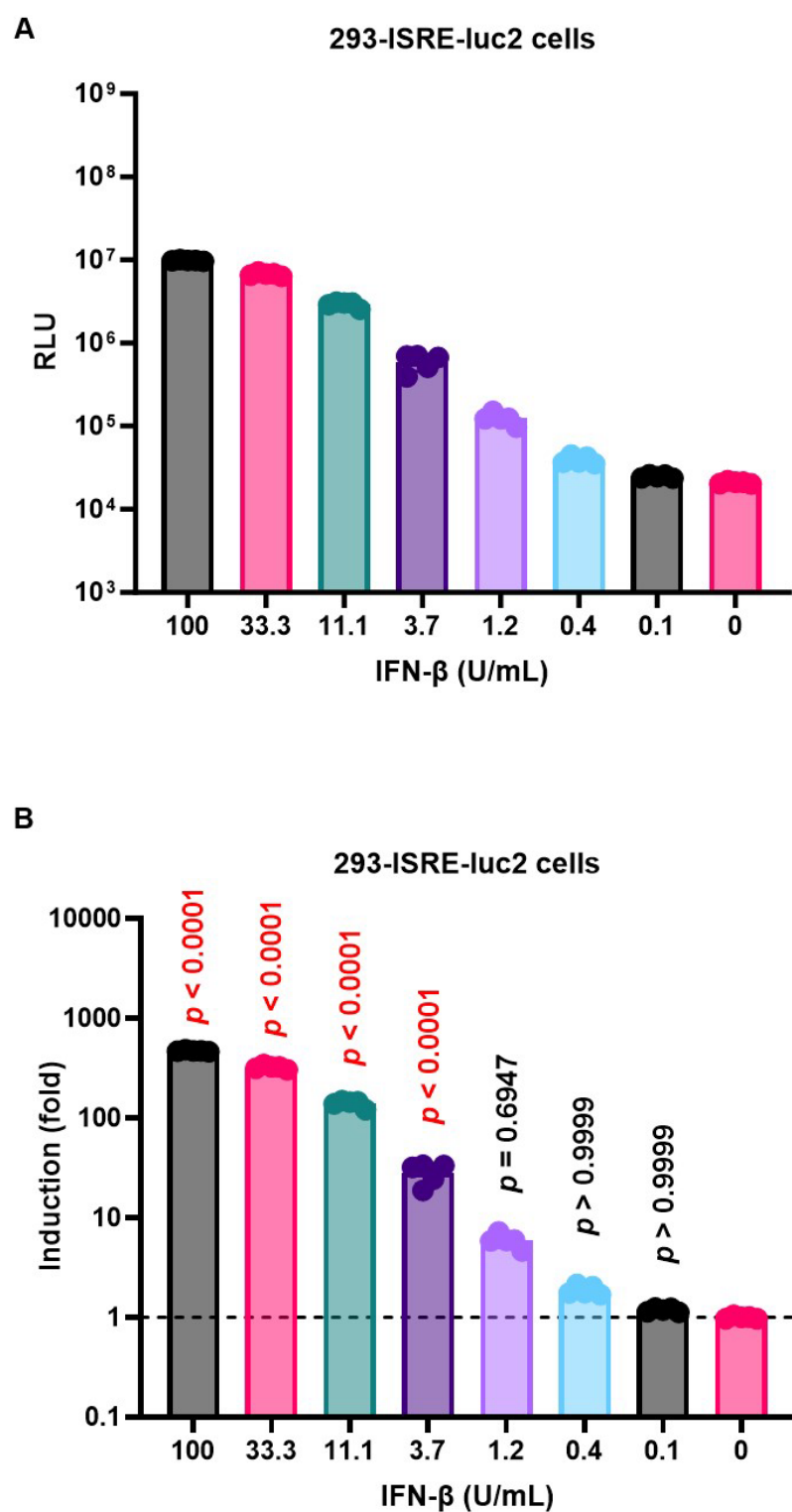
**PBHBV**



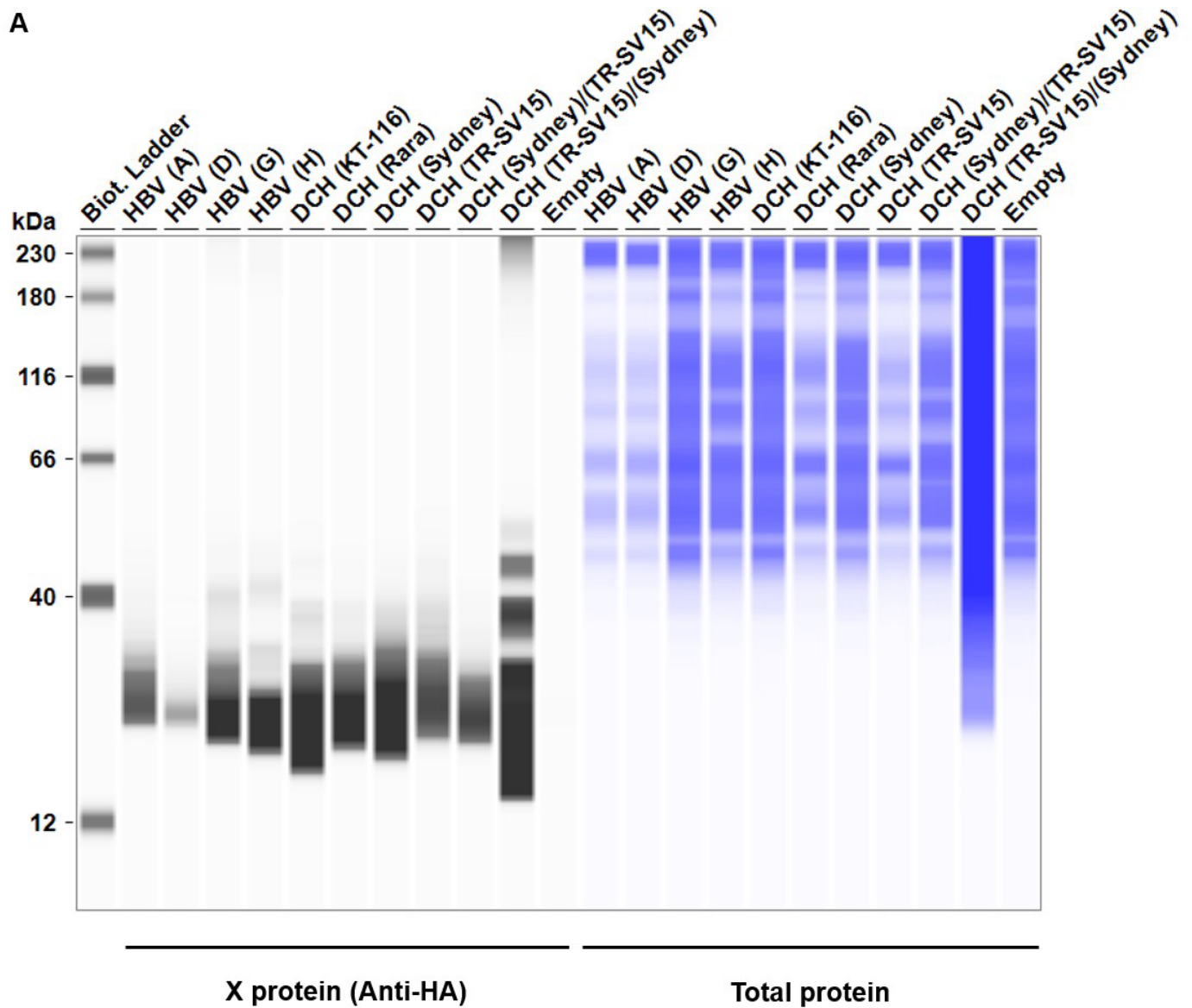




**Supplementary Figure S3. Subcellular localization of *Orthohepadnavirus* X proteins in Lenti-X 293T cells.** (A) A representative image of 40× magnification of Lenti-X 293T cells transfected with plasmids encoding *Orthohepadnavirus* X proteins. (B) A representative image of 40× magnification of Lenti-X 293T cells transfected with plasmids encoding del(1-50) or del(45-140) mutant X proteins derived from HBV (A) or DCH (KT-116). Cells were probed with Alexa Fluor 647 anti-HA.11 (orange), and nuclei are stained with NucBlue (blue).



**Supplementary Figure S4. Induction of luciferase reporter upon stimulation of 293-ISRE-luc2 cells with human IFN-β.** (A) The RLU of 293-ISRE-luc2 cells stimulated with 0-100 U/mL of human IFN-β. (B) Induction of luciferase reporter upon stimulation of 293-ISRE-luc2 cells with human IFN-β. Differences between IFN-β-treated and control cells were examined by one-way ANOVA followed by Dunnett's multiple comparison test.



Supplementary Figure S5. Expression levels of *Orthohepadnavirus* X proteins in 293T-ISRE-luc2 cells.