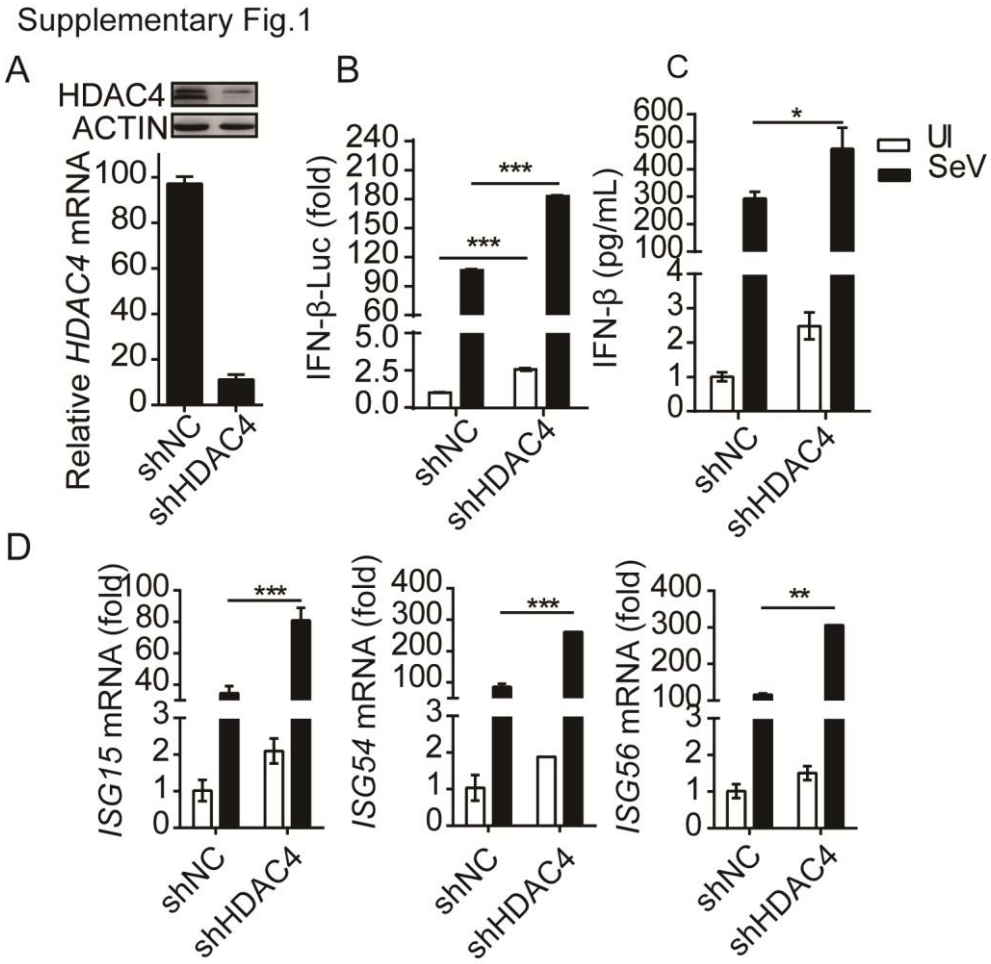
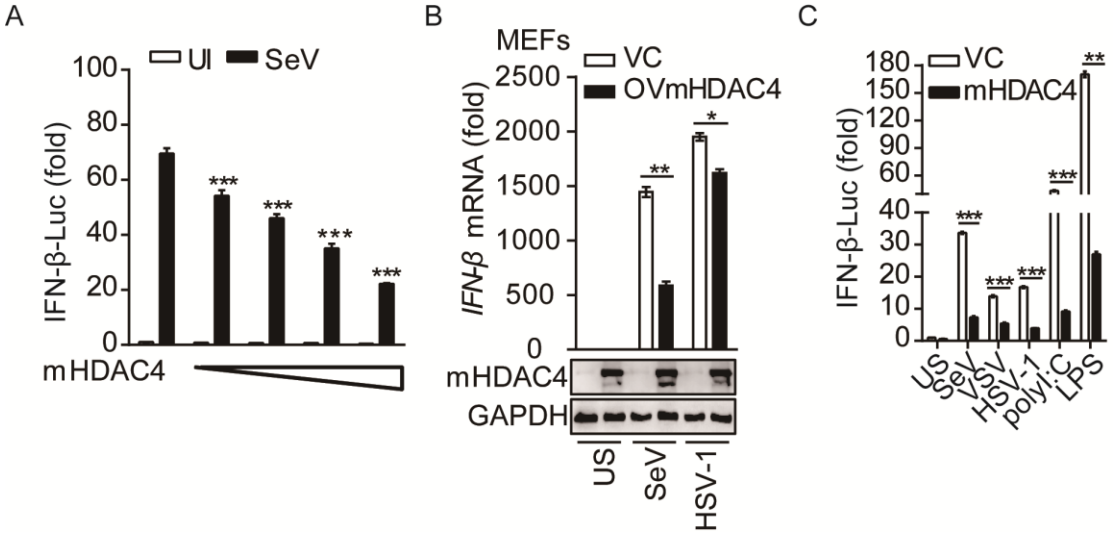
Supplementary material

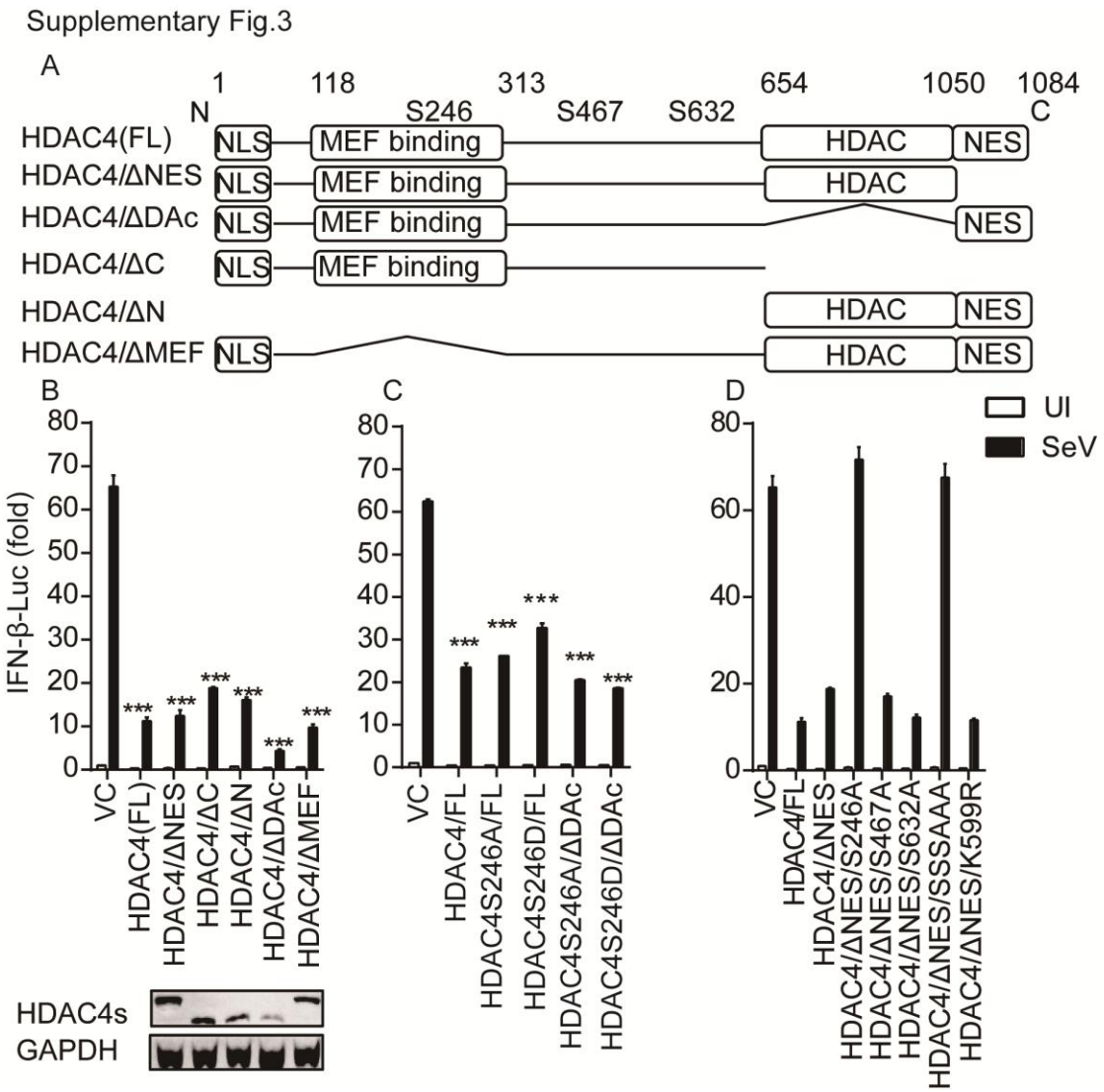


Supplementary Fig 1. Deficiency in HDAC4 arrestingly facilitates the production of IFN-β. (A) HEK293T cells were silenced with shNC or shHDAC4 to construct stable cell lines. Quantitative RT-PCR analysis of HDAC4 mRNA (below) and immunoblot analysis of HDAC4 and GAPDH (loading control throughout) (top) were carried out to analyze the Knockout efficiency. (B, C) Luciferase assays and ELISA of IFN-β (as shown in Fig 1 B, C) in shNC or shHDAC4 cell lines (2×10^5) transfected for 36 h with encoding an *IFN-β* firefly luciferase reporter (IFN-β-Luc), then left uninfected (UI) or infected for another 8 h with SeV. (D) Quantitative RT-PCR analysis of *ISG15*, *ISG54* and *ISG56* mRNA in shNC or shHDAC4 cell lines (2×10^5), then infected for another 8 h with SeV.

Supplementary Fig.2

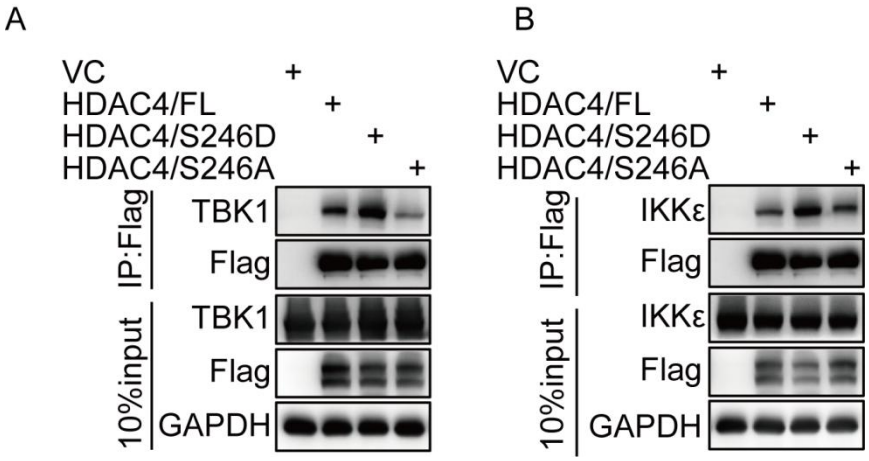


Supplementary Fig 2. HDAC4 is involved in virus-induced production of mouse IFN-β. (A) Luciferase assay analysis *IFNB1* promoter activity in MEFs infected with Lentiviral vector carrying mouse HDAC4 gene (in gradient viral load) for 1 w, then expressing with IFN-β firefly luciferase reporter (as shown in Fig 1B), left uninfected (UI) or infected for another 8 h with SeV. (B) Quantitative RT-PCR analysis of IFN-β in MEFs infected with Lentiviral vector carrying mouse HDAC4 gene for 1 w, then stimulated with SeV or HSV-1 for 8 h. (C) Luciferase assay analysis *IFNB1* promoter activity in mouse macrophages (RAW264.7) transfected with encoding an IFN-β firefly luciferase reporter treated for 8 h with SeV, VSV, HSV-1, poly(I:C) and LPS. * $p<0.05$, ** $p<0.01$ and *** $p<0.001$ (unpaired t-test). Data are from three independent experiments (a-c; mean and s.d. of three independent biological replicates per group).



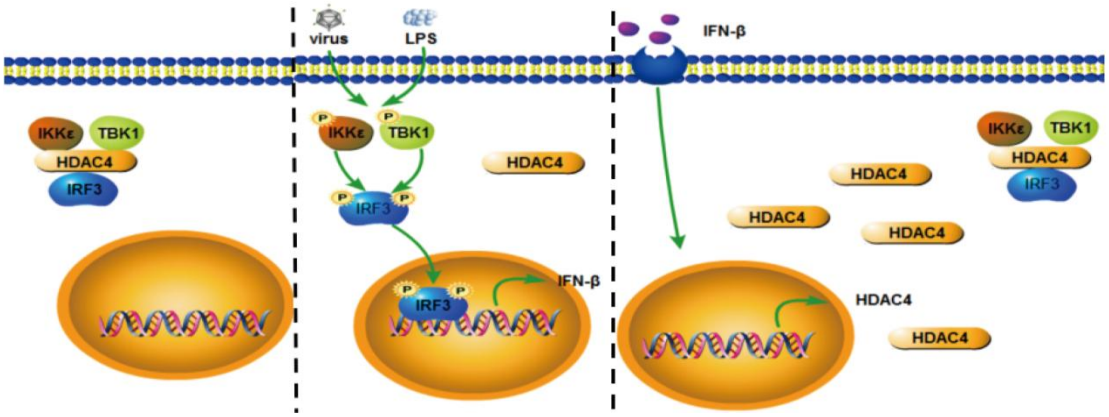
Supplementary Fig 3. Mapping the functional domain of HDAC4 in innate immunity. (A) Schematic representation of HDAC4 and its mutants. (B-D) Luciferase assay of an IFN-β reporter in HEK293T cells transfected with control vector (VC) or vector encoding to wide-type (HDAC4/FL) or mutant HDAC4 (horizontal axis) and immunoblot analysis of mutant HDAC4 containing various combinations of those domains (bottom in B).

Supplementary Fig.4



Supplementary Fig 4 (A, B) Immunoprecipitation and immunoblot analysis of lysates of HEK293T cells overexpressed HA-tagged TBK1 or HA-tagged IKKε and Flag-tagged wide-type HDAC4 (HDAC4/FL) or mutant of HDAC4 (HDAC4/S246D or HDAC4/S246A). Data are representative of three independent experiments.

Supplementary Fig.5



Supplementary Fig 5 The working patterns.

Supplementary Table S1. Primers used to amplify the different deletions of HDAC4

HDAC4 deletions	Orientation	Sequence (5'-3')
HDAC4/FL	F/R	*GCGGCCGC*CATGAGCTCCCAAAGCCATCC*AGATCT**CTA***CAGGGGCGGCTCCTC
1-1050/ΔNES	R1050	*AGATCT**CTA***GTTCTCGCAAGTCTGAGCCTCG
Δ655-1049/ΔDAc	OCF	AAGCCGAGGTTCACGACAAACGAAGAAGCCGAGACG
Δ655-1049/ΔDAc	OCR	CGTCTCGGCTTCTTCGTTTGTCGTGAACCTCGGCTTG
1-654/ΔC	R654	*AGATCT**CTA***TGTCGTGAACCTCGGCTTGG
655-1084/ΔN	F655	*GCGGCCGC*GGCCTCGTGTATGACACG
1-654-Δ118-313	OCF	ATCAAGCAACAACAGGAGAACGGTATCGCGCCCGCC
1-654-Δ118-313	OCR	GGCGGGCGCGATACCGTTCTCCTGTTGTTGCTTGATG
S246A	OCF	TCTTAGGAAAACAGCTGCTGAACCGAATCTGAAATTAC
S246A	OCR	CCGTAATTTCAGATTCGGTTCAGCAGCTGTTTTCCTAAG
S246D	OCF	TCTTAGGAAAACAGCTGATGAACCGAATCTGAAATTAC
S246D	OCR	CCGTAATTTCAGATTCGGTTCATCAGCTGTTTTCCTAAG
S467A	OCF	ACTGGGGCGGACCCAGGCGGCCCCGCTGCCCCAGAAC
S467A	OCR	GTTCTGGGGCAGCGGGGCCGCCTGGGTCCGCCCCAGTG
K559R	OCF	CAGGCCGGCGTGCAGGTGAGGCAGGAGCCCATTGAGAG
K559R	OCR	CTCAATGGGCTCCTGCCTCACCTGCACGCCGGCCTGTG
S632A	OCF	CTGTCCCGGGCGCAGGCCTCACCCGCGTCTGCCACCTTC
S632A	OCR	TGGCAGACGCGGGTGAGGCCTGCGCCCGGGACAGAG
H803L	OCF	CGCCCCCCTGGACACCTTGCGGAGGAGAGCACGCCC
H803L	OCF	CGTGCTCTCCTCCGCAAGGTGTCCAGGGGGGCGGAC
mouse-HDAC4	F/R	*AAGCTT*ATGAGCTCCCAAAGCCATCCAG*GGTACC**CTA***CAGTGGTGGTTCCTCCTCC

F indicate forward primers; R indicate reverse primers; OCF and OCR indicate overlapping complement ation forward or reverse primers, respectively. Italic indicate restriction site. Bold denote termination codon.

Supplementary Table S2. Primers for mRNA Quantification

Gene name	Orientation	Sequence (5'-3')
HDAC4	F/R	AGCGTCCGTTGGATGTCAC/CCTTCTCGTGCCACAAGTCT
IFNB1	F/R	AGGACAGGATGAACTTTGAC/TGATAGACATTAGCCAGGAG
ISG15	F/R	GAGAGGCAGCGAACTCATCTT/CCAGCATCTTCACCGTCAGG
ISG56	F/R	TAGCCAACATGTCCTCACAGAC/TCTTCTACCACTGGTTTCATGC
ISG54	F/R	GGTCTCTTCAGCATTTATTGGTG/TGCCGTAGGCTGCTCTCCA
ACTIN	F/R	GTGACGTTGACATCCGTAAAGA/GCCGGACTCATCGTACTCC

F indicate forward primers; R indicate reverse primers.