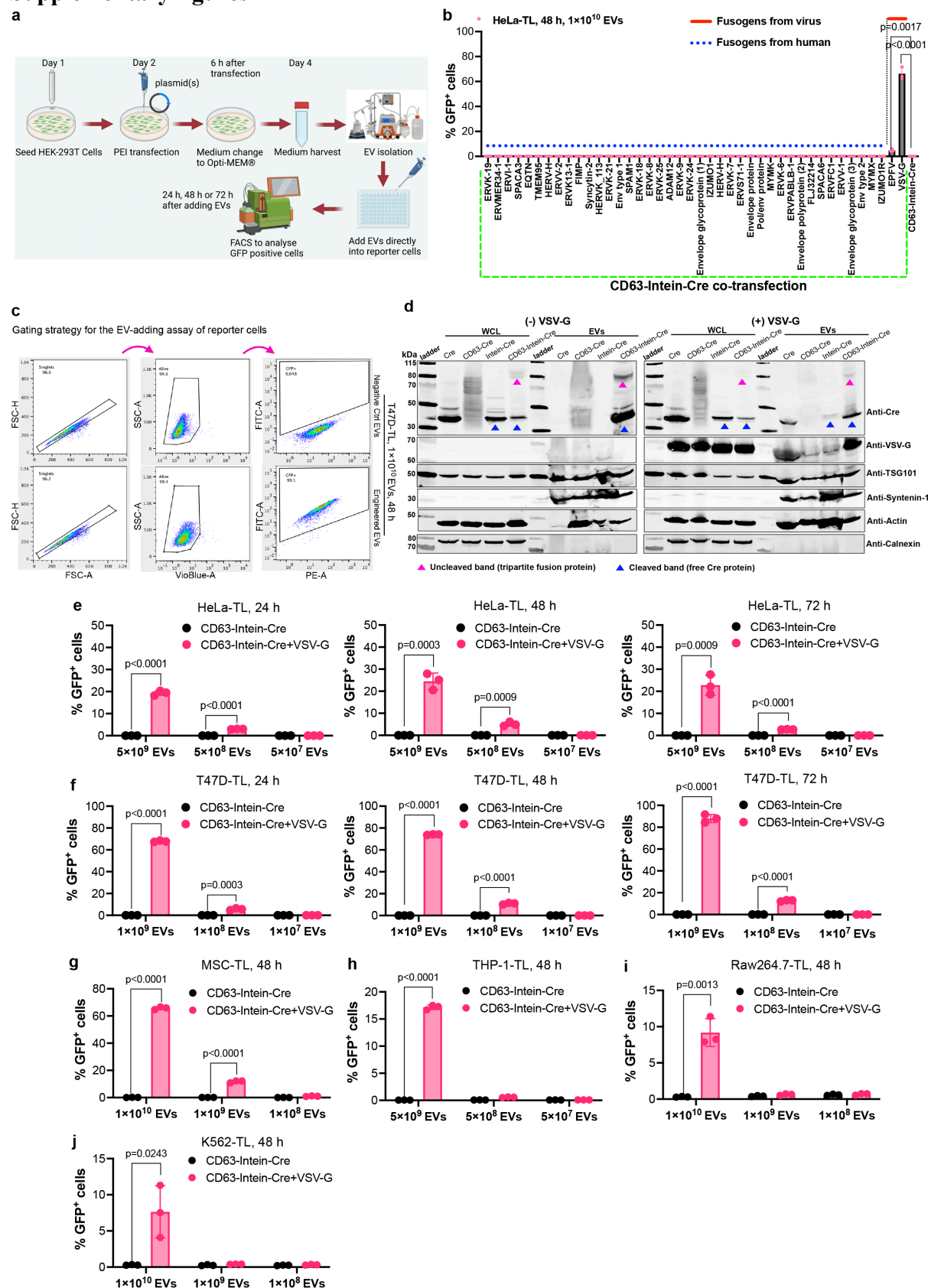
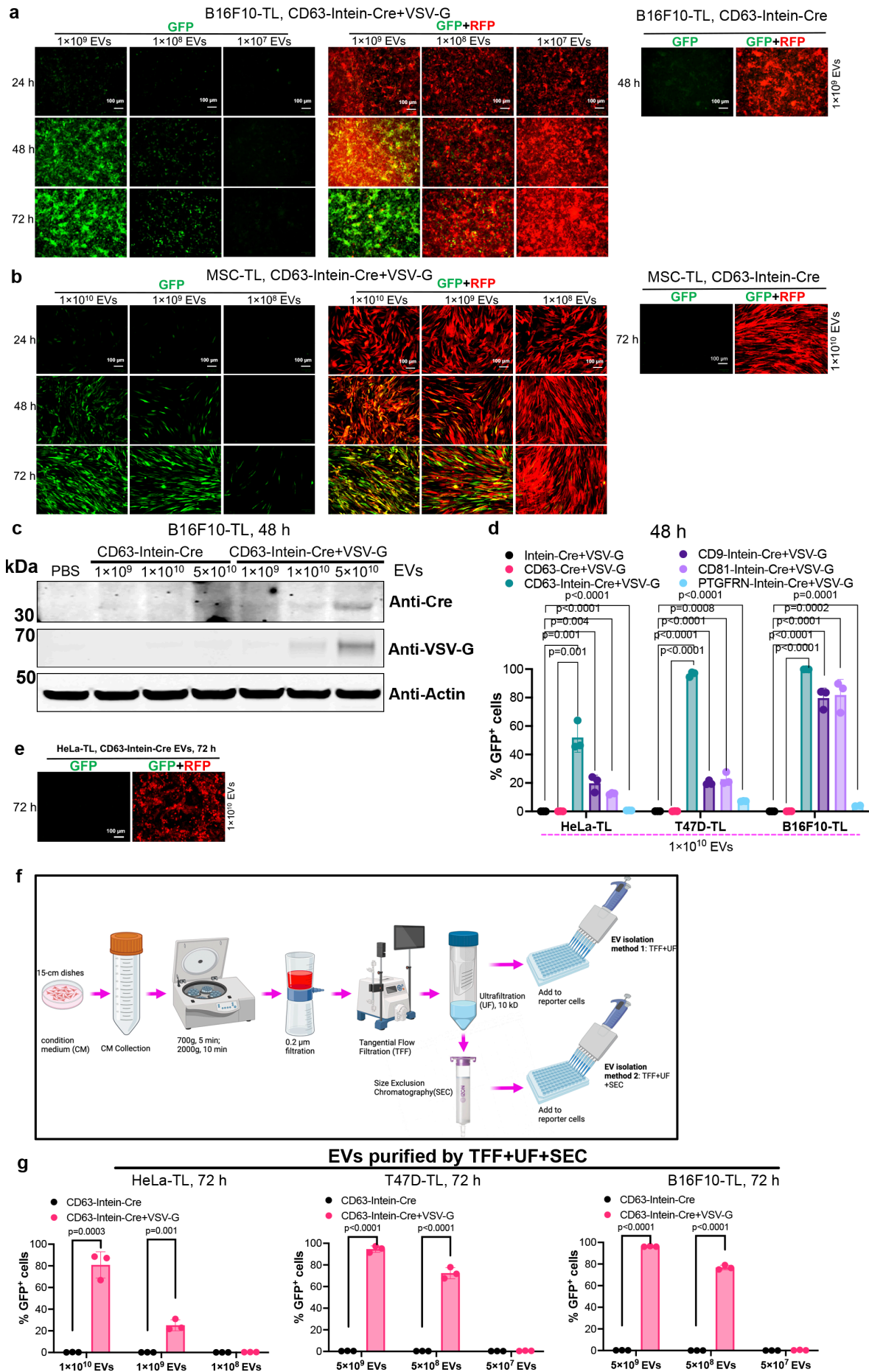
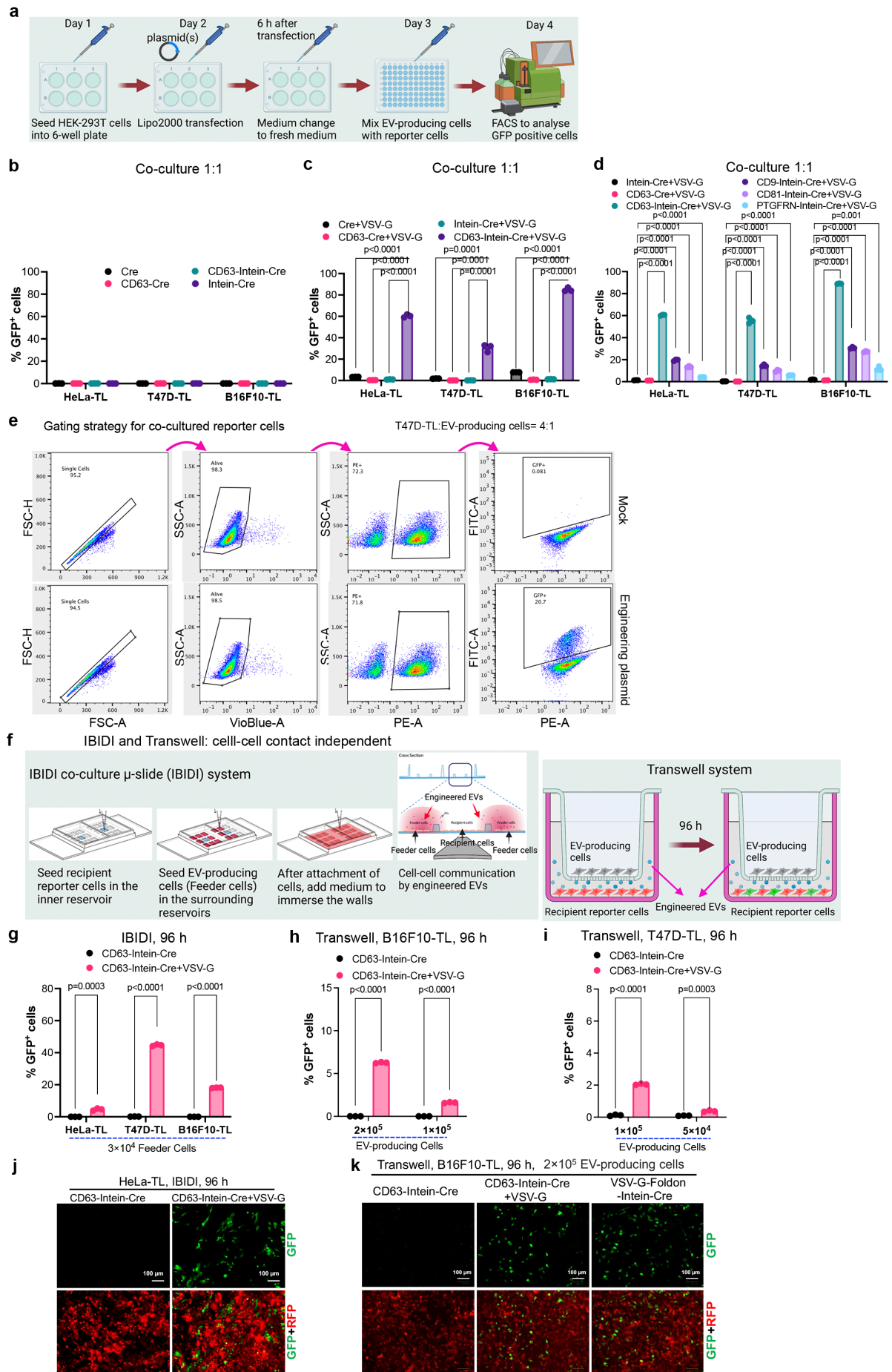




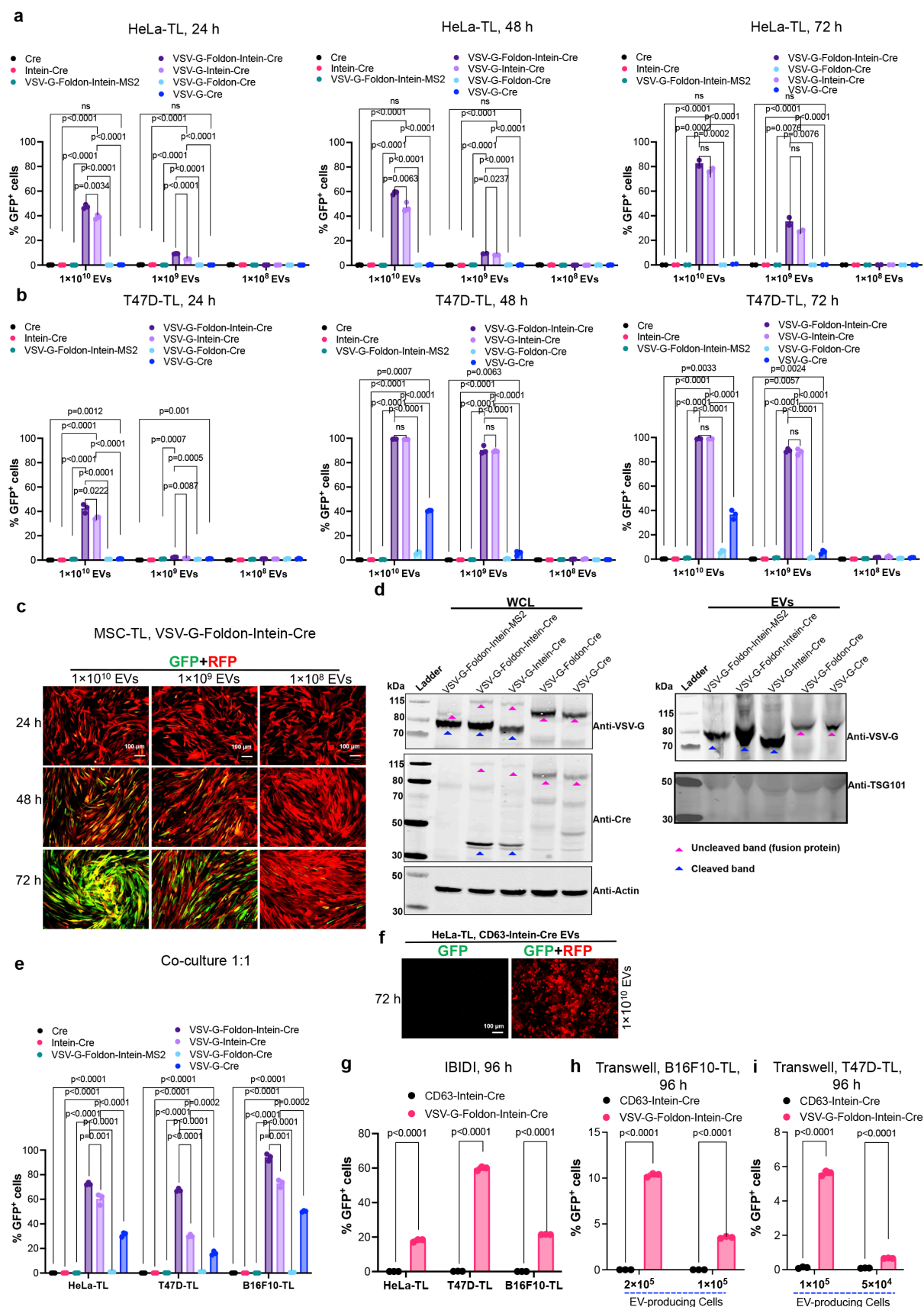
Supplementary Fig. 1. Development of VEDIC system for efficient intracellular protein delivery. **a**, A workflow schematic for the evaluation of EV-mediated Cre delivery to Cre recombinase fluorescent Traffic Light (TL) reporter cells and the subsequent analysis of GFP positive cells in recipient cells after adding isolated EVs. **b**, Fusogen screen in HeLa-TL cells after a two-day incubation with EVs. **c**, Example of the applied flow cytometry gating strategy for the analysis of GFP positive cells to evaluate Cre recombination after adding isolated EVs to fluorescent TL reporter cells. **d**, Protein expression of various engineered Cre fusion constructs in whole cell lysates (WCL) and isolated EVs derived from HEK293T cells evaluated by western blot (WB) analysis. Lysates from 5×10^5 EV-producing cells and 1×10^{10} engineered vesicles were used for the assay. TSG101, syntenin-1 and β -actin were used as EV markers and Calnexin was used as a cellular organelle marker (endoplasmic reticulum) and should be absent for EV samples. **e,f**, Percentage of GFP positive cells in HeLa-TL and T47D-TL reporter cells respectively after adding different doses of isolated engineered EVs for 1 to 3 days. **(g-j)** Percentage of recombined GFP positive cells mediated by intracellular Cre delivery through different doses of engineered EVs for 48 hours (h) in hard-to-transfect cells (**g**: MSC-TL; **h**: THP-1-TL; **i**: Raw264.7-TL; **j**: K562-TL). One-way ANOVA (Tukey) multiple comparisons test was used for analysis of (**b**); Two-way (Tukey) ANOVA multiple comparisons test was used for analysis of (**e-j**). **a** Created in BioRender.com, Zheng, W. (2025) <https://BioRender.com/s90c171>. Experiments were done with 3 biological replicates and data are shown as mean $\pm$ SD. Exact p values ($p < 0.0001$) were reported in the Source Data. Source data are provided as a Source Data file.



Supplementary Fig. 2. Fluorescent microscopy Analysis of EV-mediated Cre delivery and test of different EV-sorting domains for VEDIC system. **a,b**, Fluorescent microscopy analysis of Cre-mediated B16F10-TL and MSC-TL reporter cell activation after adding indicated doses of EVs for indicated time of incubation. Scale bar, 100 μ m, representative images. **c**, Cre and VSV-G proteins measured by WB in B16F10-TL cells after adding the indicated doses of engineered EVs in 24-well plates. **d**, Recombination in reporter cells mediated by EVs derived from engineered cells using different EV-sorting domains. **e**, Representative images showing the GFP positive HeLa-TL cells after adding EVs for 72 for CD63-Intein-Cre group (without VSV-G as negative control). Scale bar, 100 μ m. **f**, Schematic illustration for the methods used to isolate engineered EVs in this study. **g**, The percentage of recombined reporter cells using VFIC EVs isolated by TFF+UF+SEC method. Two-way ANOVA (Tukey) multiple comparisons test was used for analysis of (**d**) and (**g**). **f** Created in BioRender.com, Zheng, W. (2025) <https://BioRender.com/v11h404>. Experiments were done with 3 biological replicates and data are shown as mean $\pm$ SD. Exact p values ($p < 0.0001$) were reported in the Source Data. Source data are provided as a Source Data file.

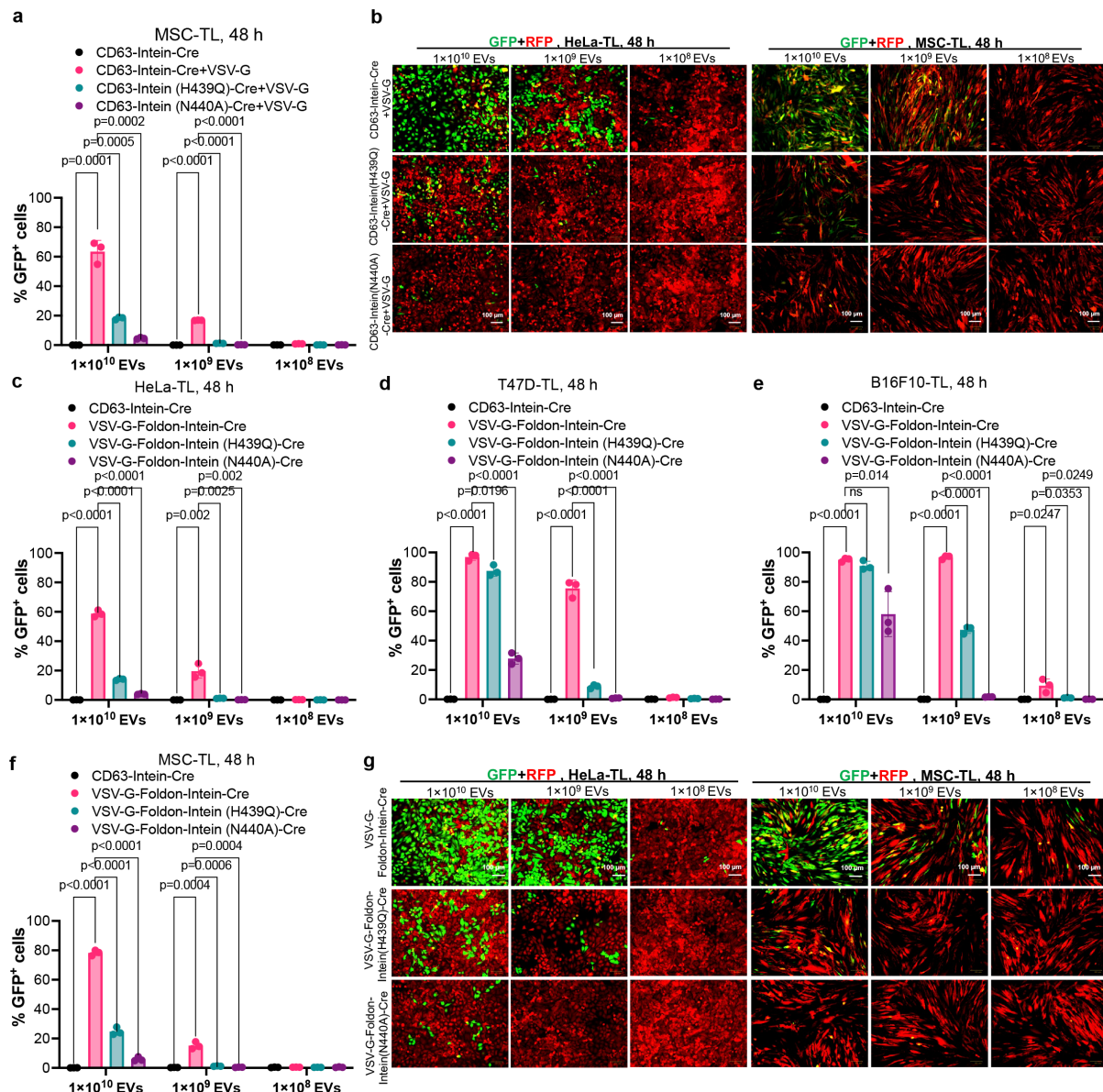


Supplementary Fig. 3. Co-culture assays for validation of the VEDIC system for efficient intracellular protein delivery. **a**, Brief workflow for the direct co-culture assay of EV-producing cells and reporter cells to show EV-mediated Cre delivery. **b**, Co-culture assay for evaluation of Cre transfer without expression of VSV-G. **c**, Analysis of Cre-mediated recombination in cells after a co-culture assay with expression of VSV-G. **d**, Comparison of Cre transfer efficiency using different EV loading domains in a direct co-culture assay. **e**, Gating strategy utilized for the analysis of Cre recombination, as indicated by the percentage of GFP positive cells, in co-culture assay. **f**, Schematic graph to show the principle and workflow of IBIDI co-culture μ -slide (IBIDI) and Transwell co-culture assays demonstrating cell-cell contact independent Cre delivery from donor cells to recipient cells. The pore size for Transwell assay was 0.4 μ m. **g**, Flow cytometry analysis of Cre delivery mediated by engineered EVs in a contact-independent co-culture assay using IBIDI assay. 3×10^4 EV-producing cells and 4×10^4 reporter cells were seeded into the surrounding reservoirs and central reservoir respectively and the analysis was performed after 4 days. **h,i**, Flow cytometry analysis of Engineered EV-mediated Cre delivery from donor cells to B16F10-TL and T47D-TL reporter cells respectively in a co-culture Transwell assay in 24-well plates. Indicated numbers of EV-producing cells were seeded into the upper chamber and the numbers of B16F10-TL and T47D-TL reporter cells in the lower chamber were 5×10^4 and 8×10^4 respectively in 24-well plate. Flow cytometry analysis was performed after 96 h. **j**, Representative fluorescence microscopy images showing Cre recombinase-induced GFP expression in HeLa-TL cells by IBIDI assay. Scale bar, 100 μ m. **k**, Fluorescence microscopy images demonstrating GFP positive cells in B16F10-TL after a Transwell assay in 24-well plates. Scale bar, 100 μ m, representative images. Two-way ANOVA (Tukey) multiple comparisons test was used for analysis of **(b-d)** and **(g-i)**. **a**, **f** Created in BioRender.com, Zheng, W. (2025) <https://BioRender.com/p83s805>, Zheng, W. (2025) <https://BioRender.com/a83q292> and Zheng, W. (2025) <https://BioRender.com/r44d413> respectively. Experiments were done with 3 biological replicates and data were shown as mean $\pm$ SD. Exact p values ($p < 0.0001$) were reported in the Source Data. Source data are provided as a Source Data file.

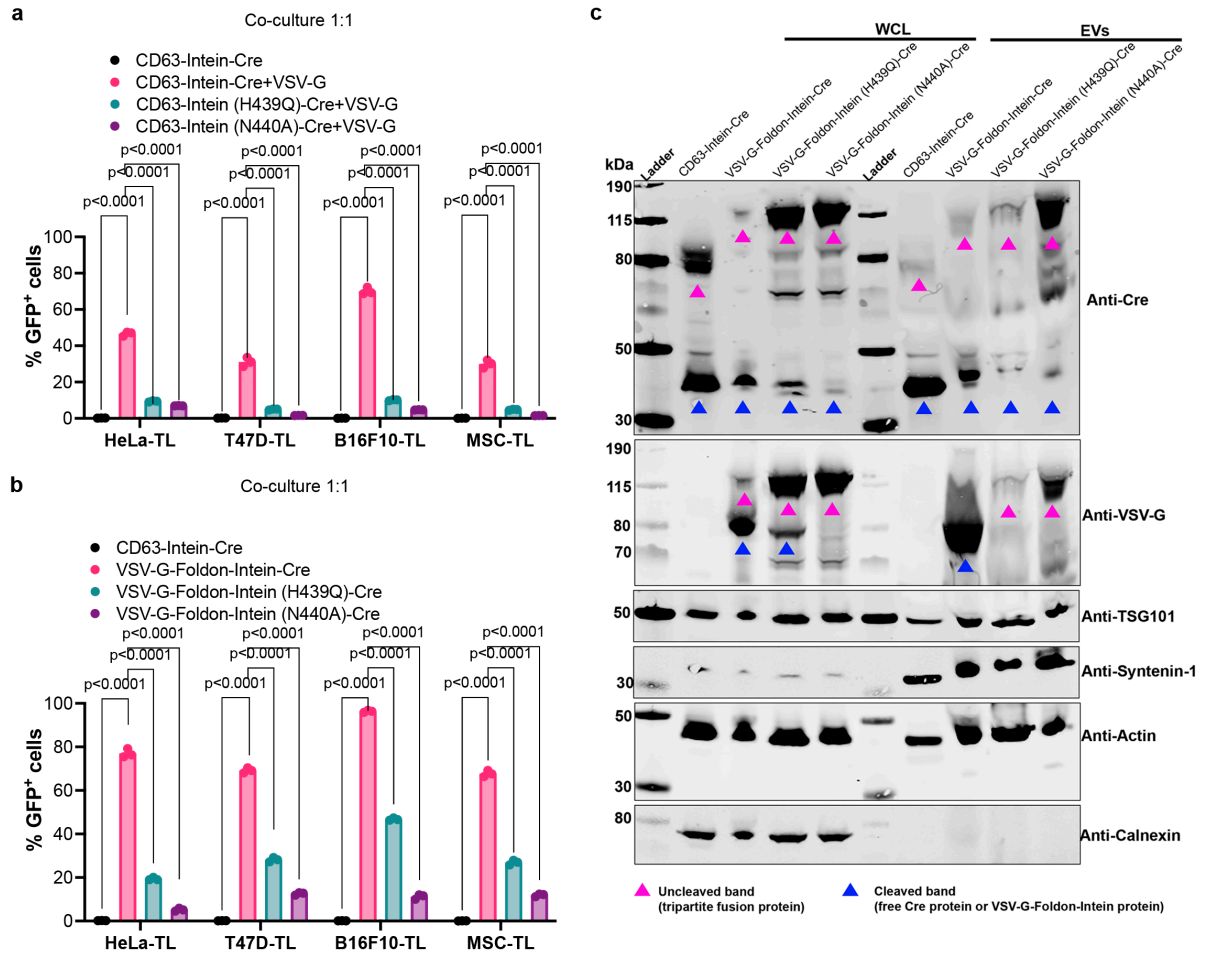


Supplementary Fig. 4. Development of VFIC system for efficient intracellular protein delivery. **a**, Percentage of GFP positive cells in HeLa-TL cells after adding engineered EVs for 1-3 days. Indicated doses of EVs were added. **b**, Percentage of recombined cells mediated by Cre from adding engineered EVs in T47D-TL cells for 1-3 days. **c**, Representative

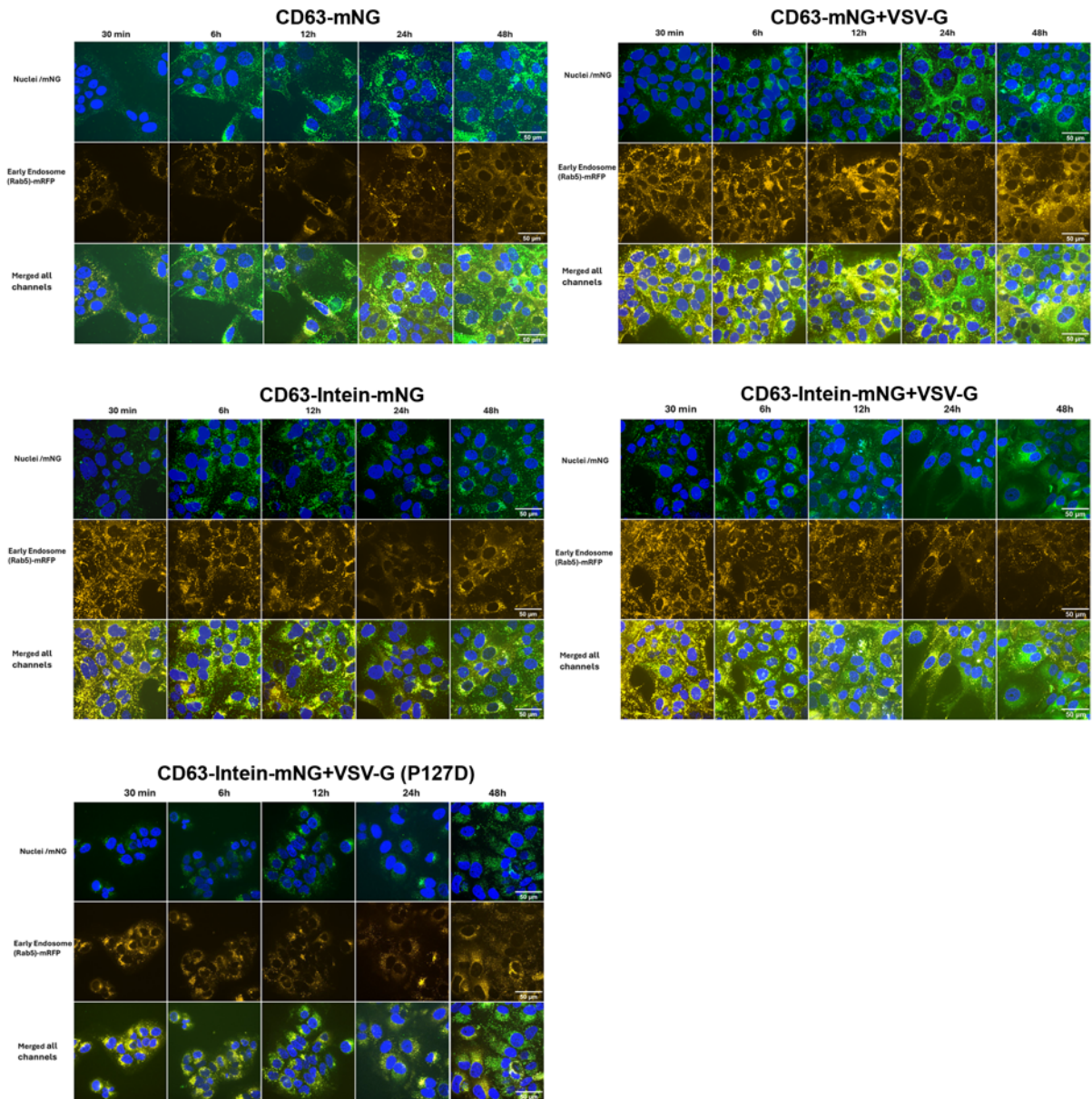
fluorescence microscopy images showed the GFP expression in MSC-TL cells after EV-mediated Cre delivery. Indicated doses of isolated EVs were incubated for the indicated time. Isolated EVs from VFIC system demonstrated dose- and time-dependent Cre delivery into recipient cells. Scale bar, 100 μm . **d**, The expression of constructs related to VFIC system as measured by WB analysis. Proteins from 5×10^5 EV-producing cells and 1×10^{10} engineered vesicles were analyzed. TSG101 was included as EV marker. **e**, Flow cytometry analysis of a direct co-culture assay using various EV engineering strategies to assay Cre transfer after 24 h. **f**, Representative images demonstrating GFP positive HeLa-TL cells after adding CD63-Intein-Cre EVs for 72 h, negative control group for VSV-G-Foldon-Intein-Cre. Scale bar, 100 μm . **g**, Percentage of GFP positive cells evaluated by IBIDI assay for 96 h. 4×10^4 EV producing cells and 4×10^4 reporter cells were seeded into the surrounding reservoirs and central reservoir respectively and flow cytometry was performed after 4 days. **h,i**, Percentage of GFP positive cells in B16F10-TL and T47D-TL cells respectively evaluated by Transwell assay for 96 h. Indicated numbers of EV-producing cells were added into the up chamber of the Transwell system and co-cultured with 5×10^4 B16F10-TL or 8×10^4 T47D-TL reporter cells in the lower chamber in 24-well plates. Two-way ANOVA (Tukey) multiple comparisons test was used for analysis of (**a** and **b**) and (**e,g-i**). Experiments were done with 3 biological replicates and data are shown as mean $\pm$ SD. Exact p values ($p < 0.0001$) were reported in the Source Data. Source data are provided as a Source Data file.



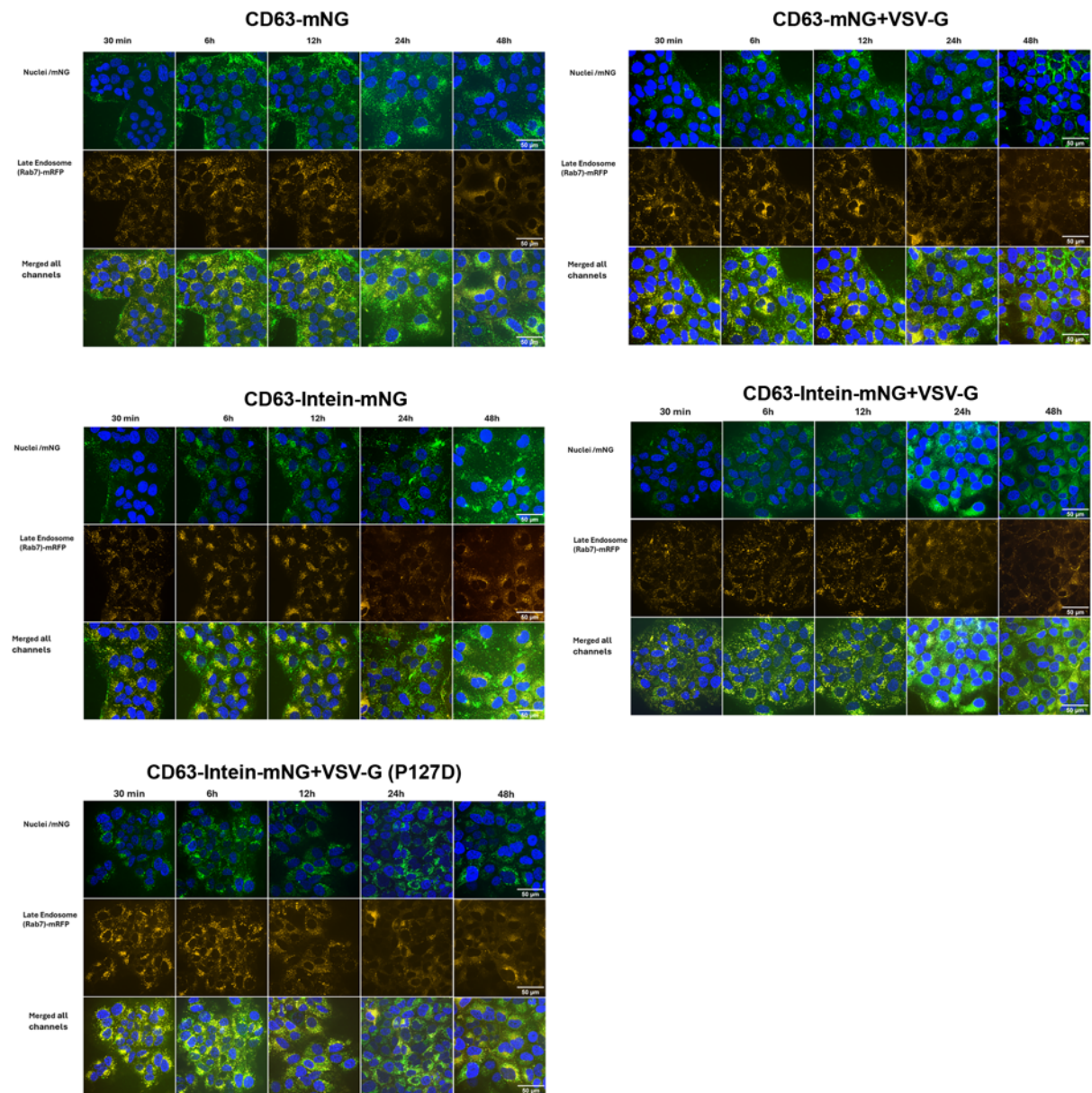
Supplementary Fig. 5. The pH-sensitive intein performs C-terminal cleavage during EV-biogenesis. **a**, Recombination in MSC-TL reporter cells mediated by EVs derived from engineered cells using mutant inteins (H439Q and N440A) in VEDIC system. **b**, Fluorescent images showing the GFP positive cells in HeLa-TL and MSC-TL cells. Scale bar, 100 μ m, representative images. **c-f**, Recombination in reporter cells (**c**: HeLa-TL; **d**: T47D-TL; **e**: B16F10-TL; **f**: MSC-TL) mediated by EVs derived from engineered cells using mutant inteins (H439Q and N440A) in VFIC system detected by flow cytometry. **g**, Representative fluorescent images for the GFP positive cells after 48 hours' adding various mutant intein variants engineered EVs at different doses for VFIC system. Scale bar, 100 μ m. Two-way ANOVA multiple comparisons test was used for analysis of (**a**) and (**c-f**). Experiments were done with 3 biological replicates and data are shown as mean $\pm$ SD. Exact p values ($p < 0.0001$) were reported in the Source Data. Source data are provided as a Source Data file.



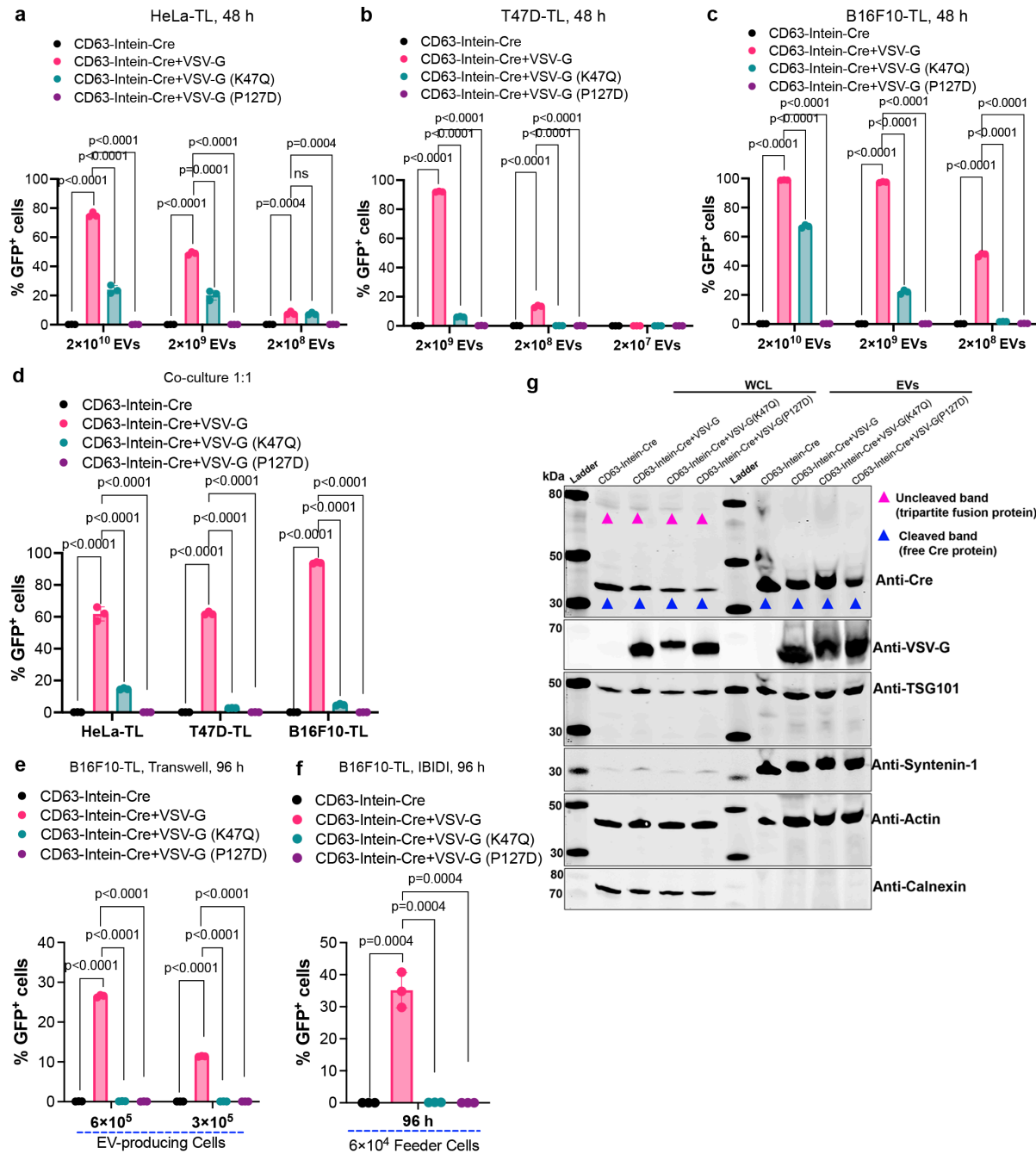
Supplementary Fig. 6. The pH-sensitive intein performs C-terminal cleavage during EV-biogenesis. **a**, Comparison of Cre transfer efficiency using different intein variants in a direct co-culture assay for VEDIC system. **b**, Flow cytometry analysis of a direct co-culture assay using intein variants to decrease Cre transfer for VFIC system. Co-culture was analyzed after 24 h incubation of the EV-producing cells and reporter cells. **c**, Protein expression of various engineered mutant intein constructs in WCL and isolated EVs derived from HEK-293T cells evaluated by WB analysis. Lysates from 5×10^5 EV-producing cells and 1×10^{10} engineered vesicles were used for the assay. TSG101, syntenin-1 and β -actin were used as EV markers and Calnexin was used as a cellular organelle marker (endoplasmic reticulum) and should be absent for EV samples. Two-way ANOVA (Tukey) multiple comparisons test was used for analysis of (**a** and **b**). Experiments were done with 3 biological replicates and data are shown as mean $\pm$ SD. Exact p values ($p < 0.0001$) were reported in the Source Data. Source data are provided as a Source Data file.



Supplementary Fig. 7. Co-localization of early endosome marker (Rab5) with the mNG in Huh7 cells at different time points (dynamic change) after adding engineered EVs. Rab5 was co-localized with mNG in different engineering groups at different time points.

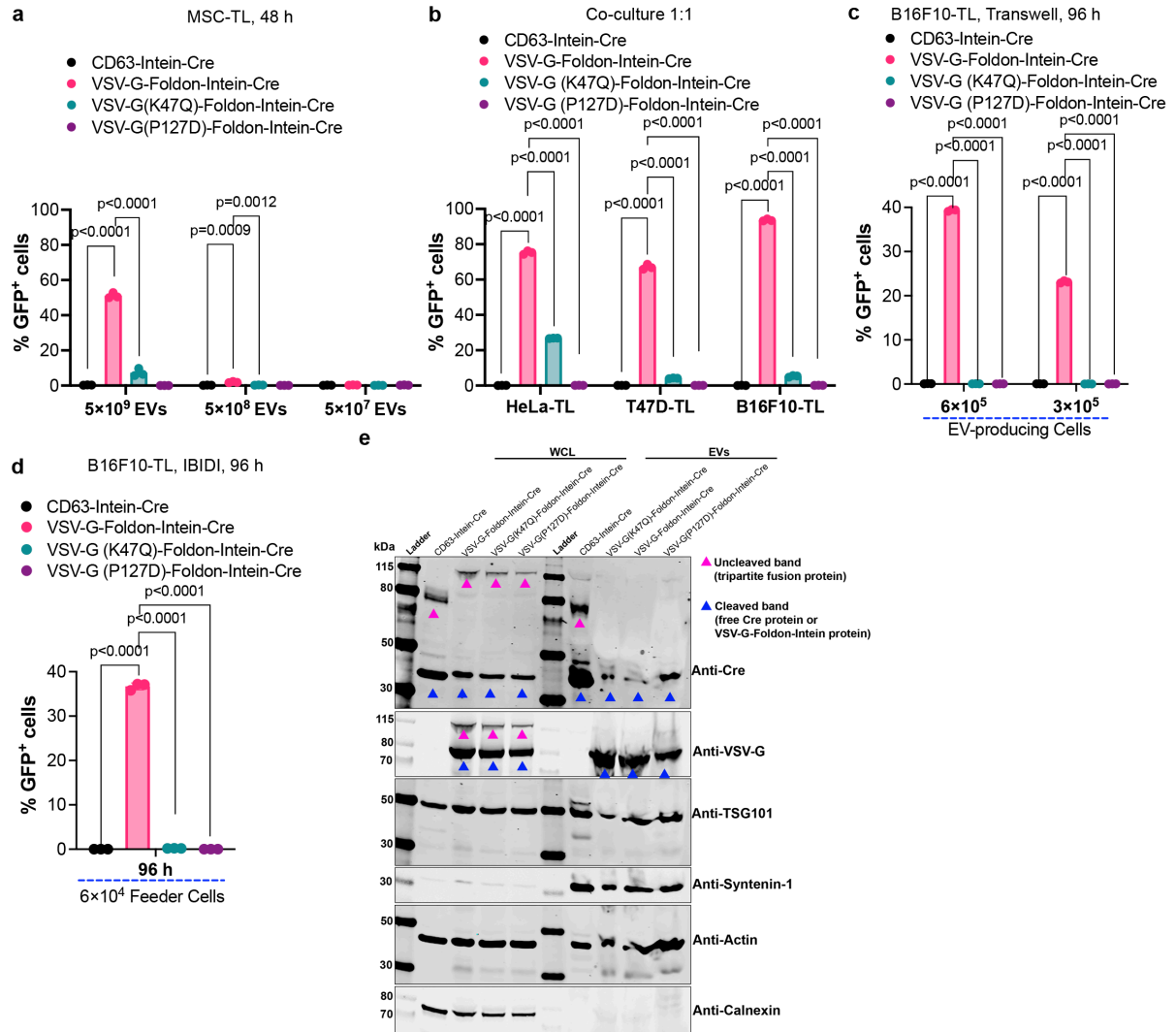


Supplementary Fig. 8. Co-localization of late endosome marker (Rab7) with the mNG in Huh7 cells at different time points (dynamic change) after adding engineered EVs. Rab7 was co-localized with mNG in different engineering groups at indicated time points.



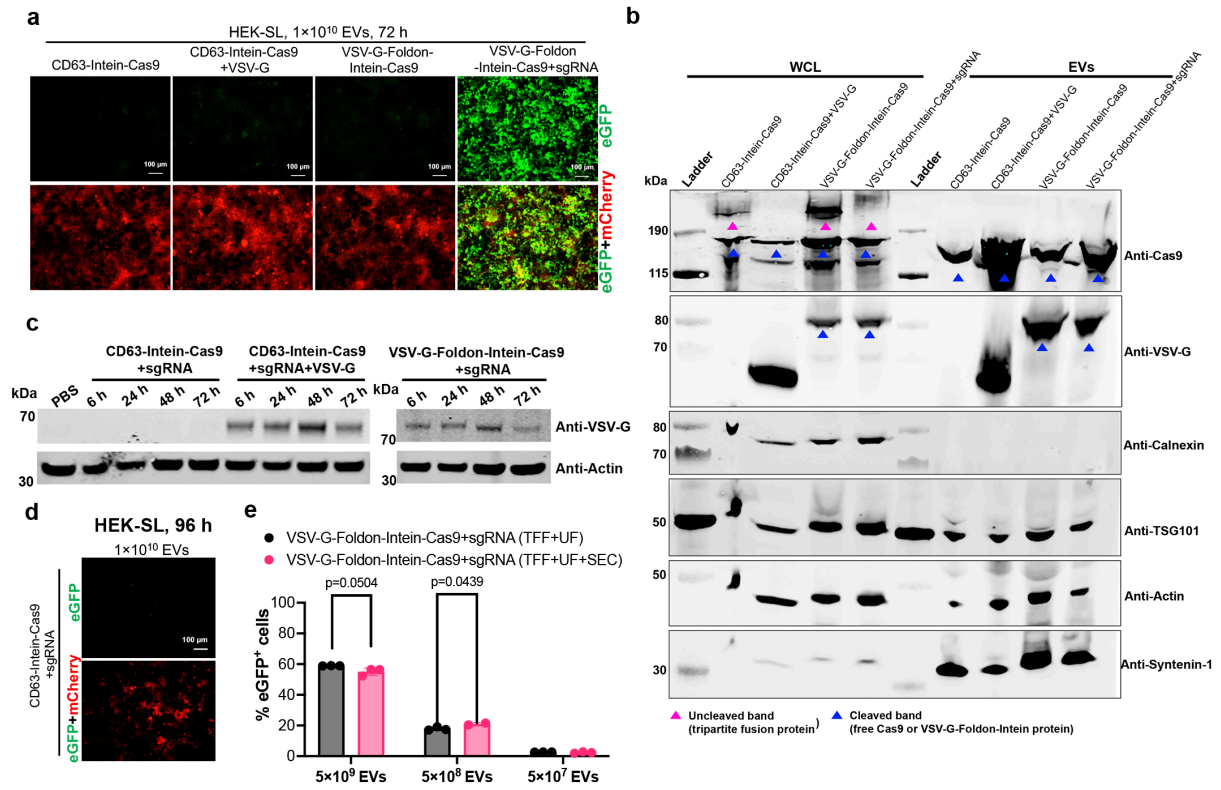
Supplementary Fig. 9. VSV-G boosts endosomal escape following receptor-mediated endocytosis of engineered EVs into recipient cells. **a-c**, Percentage of GFP positive HeLa-TL, T47D-TL and B16F10-TL cells after adding CD63-Intein-Cre EVs or wild type, P127D or K47Q VSV-G plus CD63-Intein-Cre EVs, as evaluated by flow cytometry. **d**, Direct co-culture assay to show the recombination efficiency of different engineered EVs. VSV-G (K47Q) and VSV-G (P127D) mutants were expressed in the EV-producing cells and resulting in abolished Cre delivery. **e**, Percentage of recombined GFP positive cells after a Transwell assay with B16F10-TL cells. VSV-G (K47Q) and VSV-G (P127D) mutants were expressed in the EV producing cells. Indicated numbers of EV-producing cells and 5×10^4 B16F10-TL cells were used for the assay in 24-well plates. **f**, Percentage of GFP positive cells evaluated by IBIDI assay in B16F10-TL cells. Indicated number of EV-producing cells and 4×10^4 reporter cells were used for this assay. **g**, Protein expression of mutated VSV-G-related constructs both in WCL of and isolated EVs evaluated by WB analysis. Proteins from 5×10^5 EV producing cells and 1×10^{10} engineered vesicles were used for the assay. TSG101, syntenin-1 and β -actin

were used as EV markers and Calnexin was used as cellular organelle marker (endoplasmic reticulum). Two-way ANOVA (Tukey) multiple comparisons test was used for analysis of (a-f). Experiments were done with 3 biological replicates and data are shown as mean $\pm$ SD. Exact p values ($p < 0.0001$) were reported in the Source Data. Source data are provided as a Source Data file.

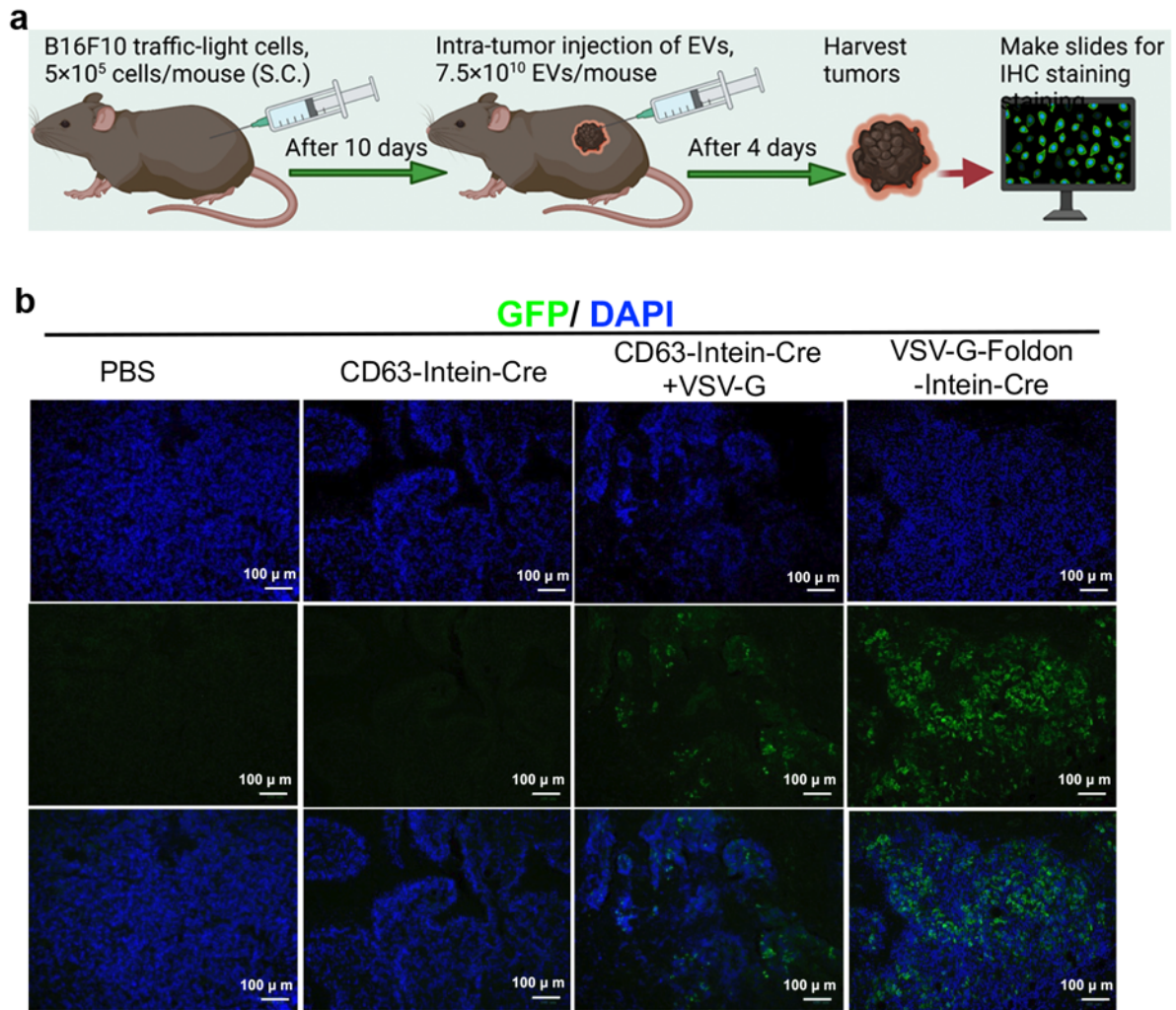


Supplementary Fig. 10. VSV-G boosts endosomal escape following receptor-mediated endocytosis of engineered EVs into recipient cells. a, Percentage of GFP positive MSC-TL cells after adding wild type, P127D or K47Q VSV-G directly fused with Foldon-Intein-Cre EVs, as evaluated by flow cytometry. **b**, Direct co-culture assay to show the recombination efficiency of different engineered EVs. VSV-G (K47Q) and VSV-G (P127D) mutants were expressed in the EV-producing cells and resulting in abolished Cre delivery. **c**, Percentage of recombined GFP positive cells after a Transwell assay with B16F10-TL cells. VSV-G (K47Q) and VSV-G (P127D) mutants were expressed in the EV producing cells. Indicated numbers of EV-producing cells and 5 $\times$ 10⁴ B16F10-TL cells were used for the assay in 24-well plates. **d**, Percentage of GFP positive cells evaluated by IBIDI assay in B16F10-TL cells. Indicated number of EV-producing cells and 4 $\times$ 10⁴ reporter cells were used for this assay. **e**, Protein expression of directly fused mutated VSV-G-related constructs both in WCL of and isolated EVs evaluated by WB analysis. Proteins from 5 $\times$ 10⁵ EV producing cells and 1 $\times$ 10¹⁰ engineered vesicles were used for the assay. TSG101, syntenin-1 and β -actin were used as EV markers and Calnexin was used as cellular organelle marker (endoplasmic reticulum). Two-way

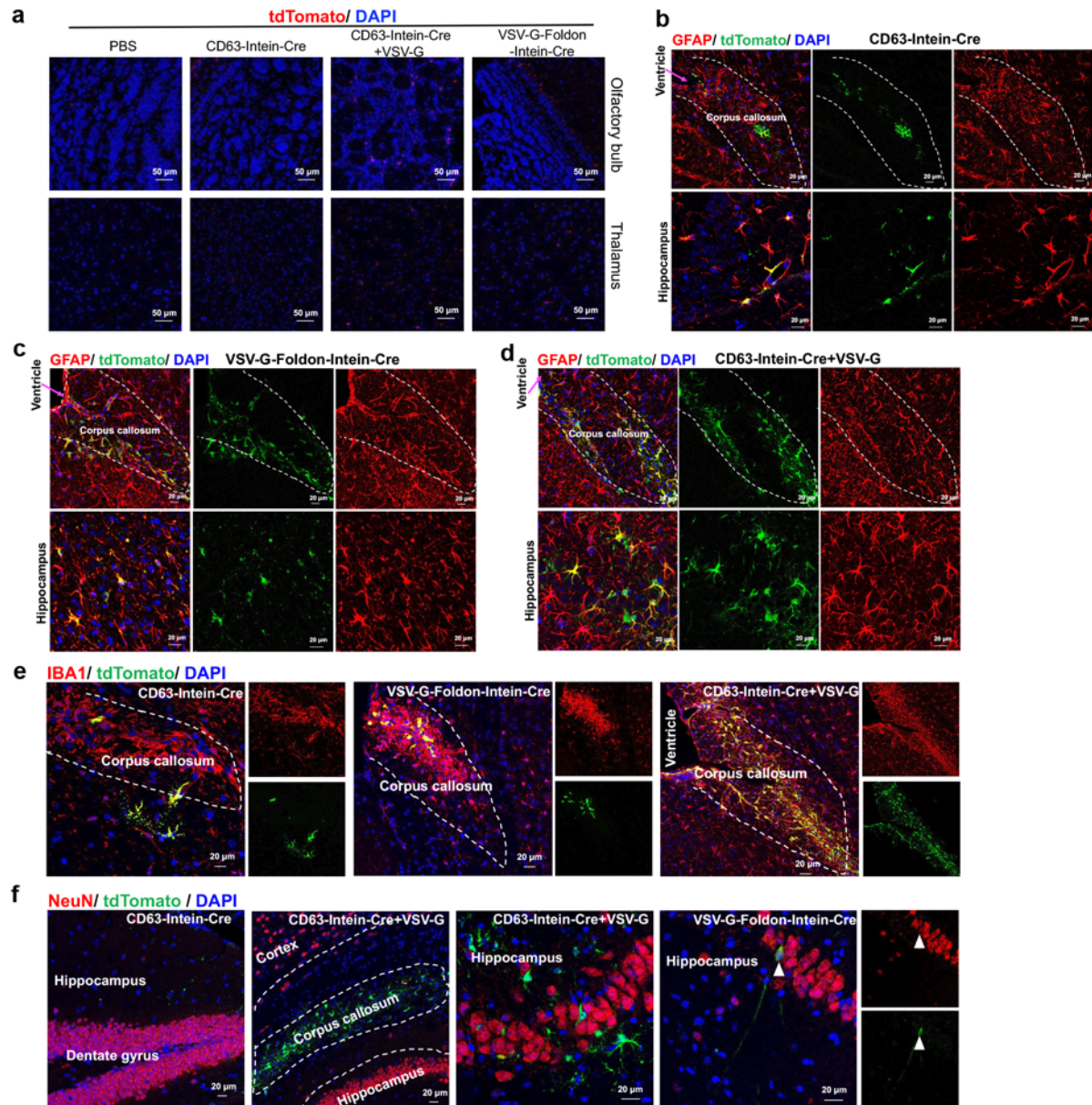
ANOVA (Tukey) multiple comparisons test was used for analysis of (a-d). Experiments were done with 3 biological replicates and data are shown as mean±SD. Exact p values ($p < 0.0001$) were reported in the Source Data. Source data are provided as a Source Data file.



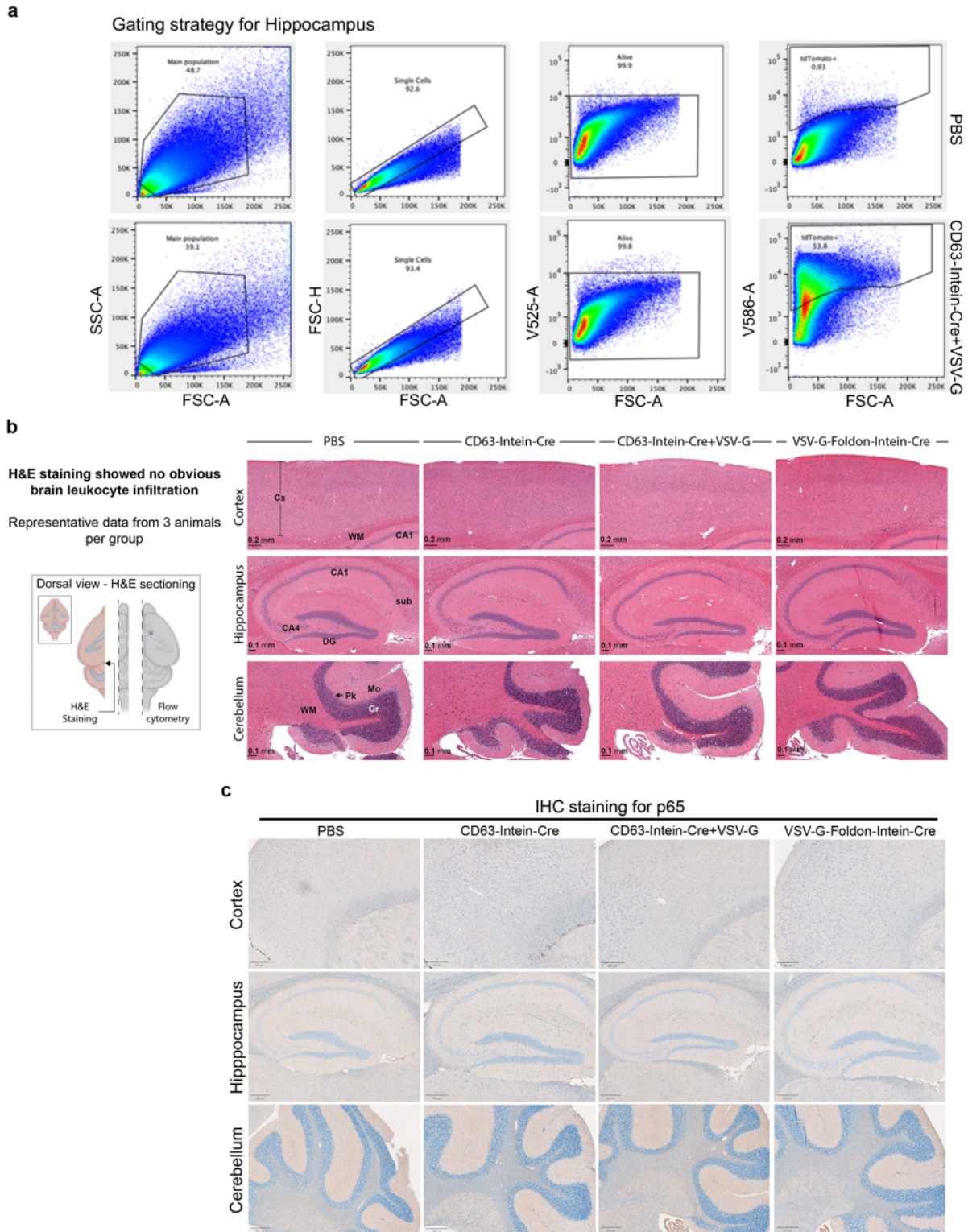
Supplementary Fig. 11. Characterization of Cas9-mediated genome editing in reporter cells by engineered EVs. **a**, Fluorescence microscopy images demonstrating functional Cas9/sgRNA RNPs delivery, as indicated by GFP positive signals, in HEK293T stoplight (HEK-SL) reporter cells after addition of indicated doses of EVs for 72 h. Scale bar, 100 μ m, representative images. **b**, Protein expression of Cas9 related constructs in both whole cell lysate (WCL) and EVs evaluated by western blot analysis. Proteins from 5×10^5 EV-producing cells and 1×10^{10} engineered vesicles were used for the assay. TSG101, syntenin-1 and β -actin were used as EV markers while Calnexin was used as cellular organelle marker (endoplasmic reticulum). **c**, Dynamic changes of VSV-G protein from different EV groups in HEK-SL cells determined by western blot analysis. **d**, eGFP positive HEK-SL cells after adding CD63-Intein-Cas9+sgRNA EVs (without VSV-G as the negative control). Representative images, scale bar, 100 μ m. **e**, Direct comparison of the editing efficiency of VFIC EVs in HEK-SL cells (96 h) isolated by TFF+UF and TFF+UF+SEC respectively. Two-way ANOVA (Tukey) multiple comparisons test was used for analysis of (e). Experiments were done with 3 biological replicates except the VSV-G-Foldon-Intein-Cas9+sgRNA (TFF+UF+SEC) 5×10^8 EV group which had 2 biological replicates for (e) and data are shown as mean±SD. Exact statistical analysis was reported in the Source Data. Source data are provided as a Source Data file.



Supplementary Fig. 12. Cre recombination in melanoma-xenograft. a, Workflow for the intratumoral injection model. **b,** Representative immunofluorescence images from tumor tissues after intratumoral injection of engineered EVs to induce GFP expression. Scale bar, 100 μ m. n=3 mice per group. **a** Created in BioRender.com, Zheng, W. (2025) <https://BioRender.com/o78k534>.

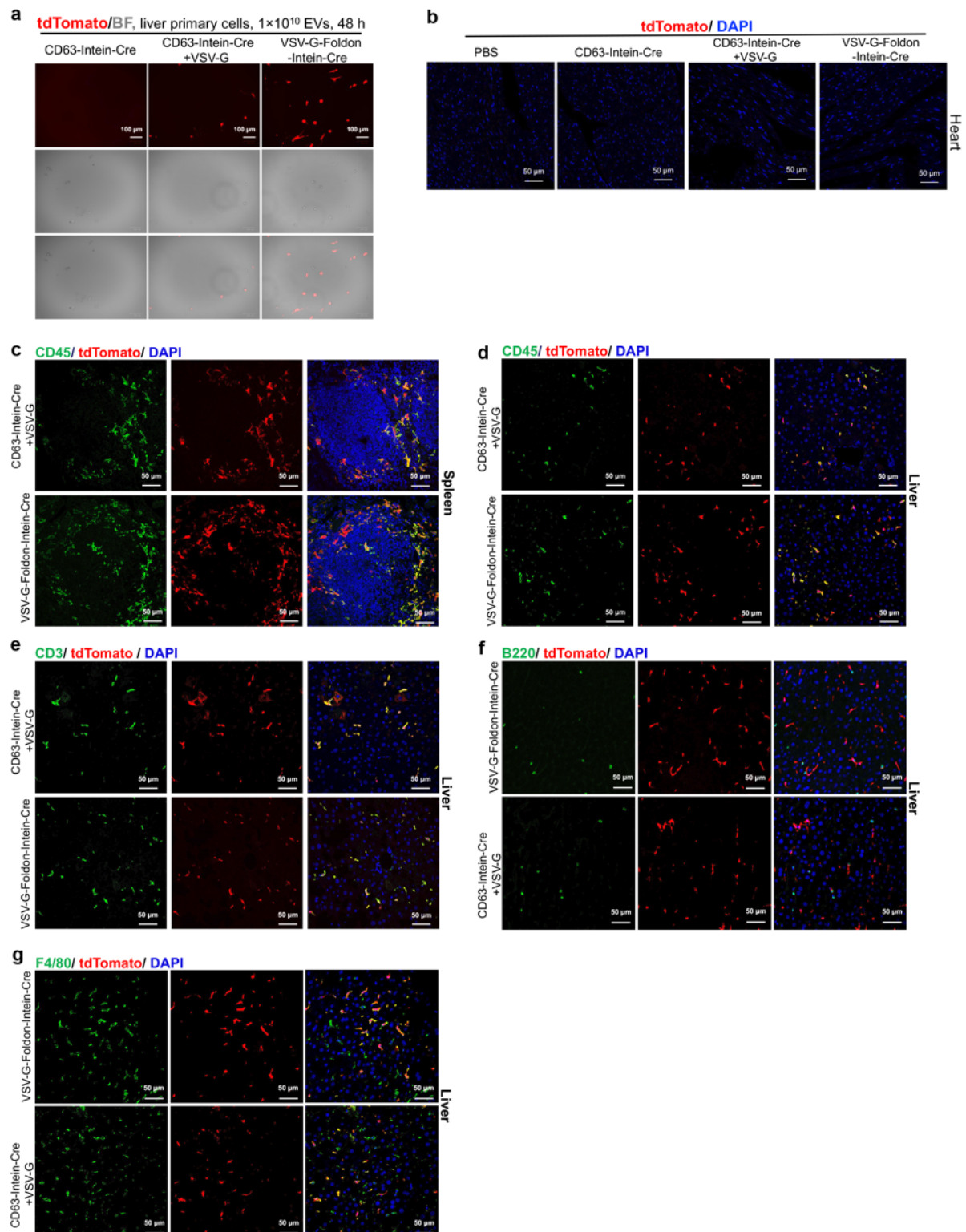


Supplementary Fig. 13. Cre-mediated recombination in R26-LSL-tdTomato reporter mice by VEDIC and VFIC systems after local injection. **a**, IHC staining of the olfactory bulb and thalamus one week after ICV injection of the engineered EVs. Scale bar, 50 μ m. **b-d**, Co-staining of tdTomato with the astrocyte marker GFAP in corpus callosum (outlined by dashed lines) and hippocampus one week after ICV injection of engineered EVs. Scale bar, 20 μ m. **e**, Co-staining of microglia marker IBA1 with tdTomato in corpus callosum (highlighted by dashed lines) after ICV injection of engineered EVs. Scale bar, 20 μ m. **f**, Co-staining of neuron cell marker NeuN with tdTomato in different regions of brain after ICV injection of different engineered EVs. Scale bar, 20 μ m. White arrow indicates sporadic cells with co-localization of NeuN and RFP. n=3 mice for PBS and n=4 for other groups, representative images for (a-f).



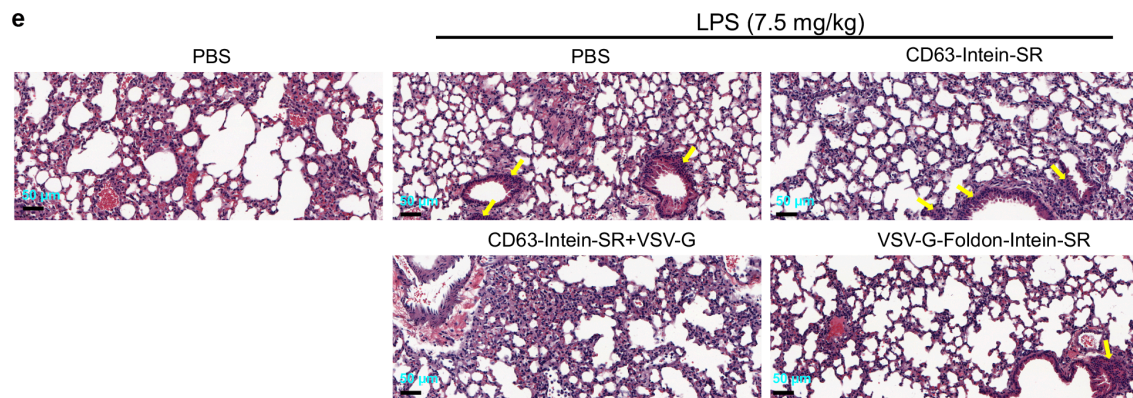
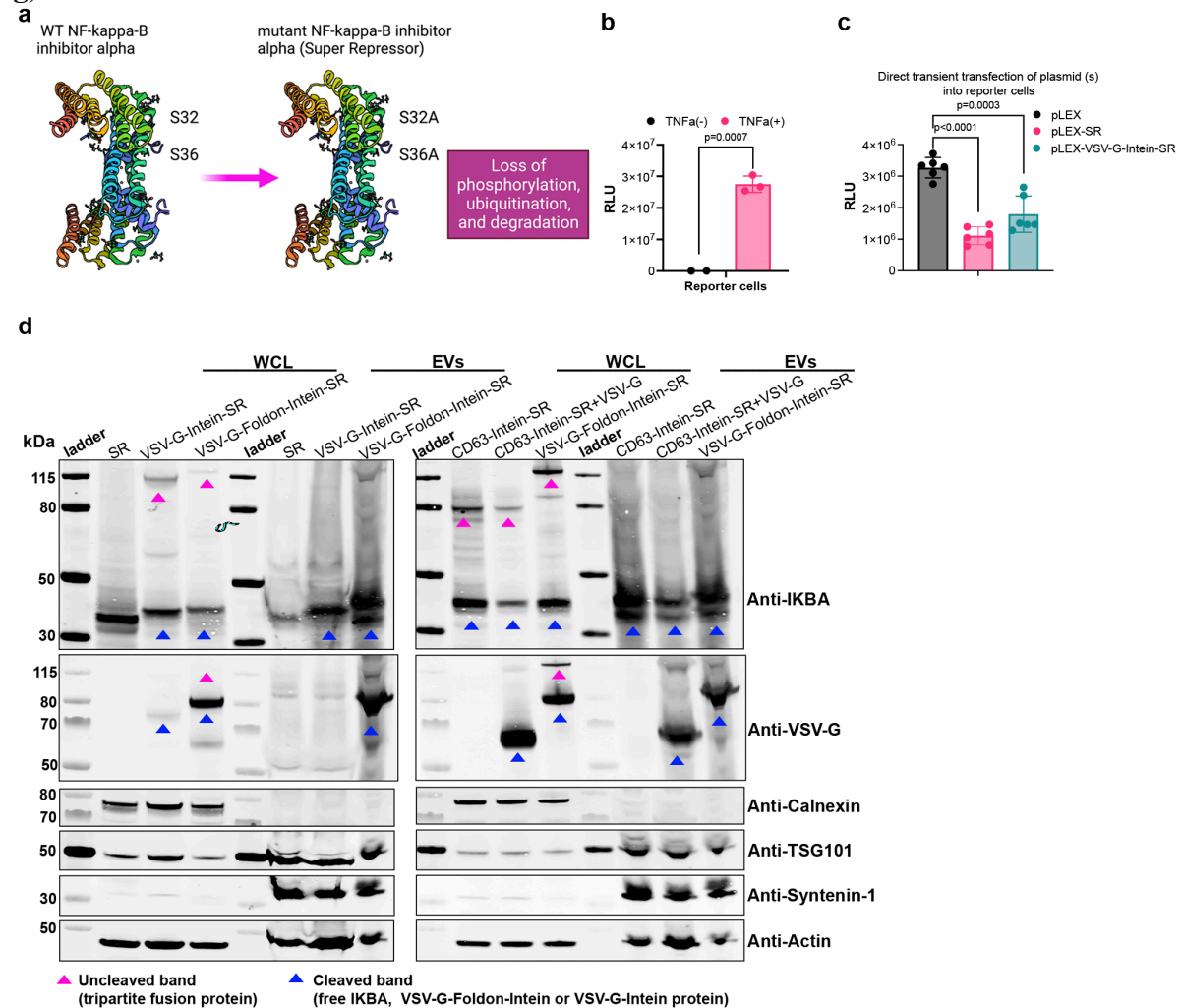
Supplementary Fig. 14. Gating strategy of FACS for evaluating tdTomato positive cells, H&E and IHC staining of p65 from different regions of brain after minipump ICV injections. **a**, Representative gating strategy of FACS to measure the tdTomato positive cells from different regions of brain after minipump ICV injections. **b**, H&E staining to show the infiltration of inflammatory cells in different regions of brain after minipump ICV injections. Scale bar, 200 μ m for cortex and 100 μ m for hippocampus and cerebellum, representative images. Hematoxylin-Eosin (H&E) for brain sagittal sections. (Cx) cortex; (WM) white matter; Ammon's horn divided in CA1- CA4 regions; (DG) dentate gyrus; (sub) subiculum; (Mo) molecular layer; (Pk) Purkinje cell layer and (Gr) granular layer. **c**, IHC staining of p65 to demonstrate

activation of inflammation in different regions of brain after minipump ICV injections. Scale bar, 250 μ m, representative images.



Supplementary Fig. 15. Cre recombination in R26-LSL-tdTomato reporter mice following systemic VEDIC and VFIC EV-mediated Cre delivery. **a**, Representative fluorescent microscopic images demonstrated tdTomato positive liver primary cells from R26-LSL-tdTomato reporter mice after adding VEDIC and VFIC EVs for 2 days. Scale bar, 100 μ m. **b**, IHC staining of the heart, one week after IP injection of the engineered EVs. Scale bar,

50 μm . **c**, Co-staining of tdTomato with the general leukocyte marker CD45 in spleen as detected by immunofluorescence one week after IP injection of engineered EVs. Scale bar, 50 μm . **d**, Co-staining of general leukocyte marker CD45 with tdTomato expression in liver after IP injection of engineered EVs. Scale bar, 50 μm . **e**, C-localization of T cell marker CD3 with tdTomato in liver one week after IP injection of engineered EVs as detected by IHC co-staining. Scale bar, 50 μm . **f**, Co-localization of B cell marker B220 with tdTomato in liver one week after IP injection of different engineered EVs. Scale bar, 50 μm . **g**, Co-expression of macrophage marker F4/80 with tdTomato in liver evaluated by IHC co-staining one week after injecting engineered EVs. Scale bar, 50 μm . $n=3$ mice per group, representative images for (b-g).



Supplementary Fig. 16. The design of super repressor (SR) inhibitor of NF- κ B constructs and their expression and function in vitro. **a**, The mutations and properties of the super repressor inhibitor of NF- κ B. **b**, Activation of luciferase expression 6 h after TNF- α stimulation (10 ng/ml) in HEK-Blue-NF- κ B reporter cells. **c**, Significant decrease of the luciferase signals from lysate of HEK-Blue-NF- κ B reporter cells after transient transfection of SR-related constructs. 48 h after transfection cells were stimulated with TNF- α for 6 h prior to luciferase detection. **d**, Protein expression of SR-related constructs in both WCL and isolated EVs evaluated by WB analysis. Proteins from 5×10^5 EV-producing cells and 1×10^{10} engineered vesicles were used for the assay. TSG101, syntenin-1 and β -actin were used as EV markers while Calnexin was used as cellular organelle marker (endoplasmic reticulum). **e**, Infiltration of inflammatory cells in lung tissues as shown by the representative histology images (yellow arrows indicate the inflammatory cells). Scale bar, 50 μ m. Two-tailed T-test was used for the analysis of (**b**); One-way ANOVA (Tukey) multiple comparisons test was used for analysis of (**c**). **a** Created in BioRender.com, Zheng, W. (2025) <https://BioRender.com/z26t488>. Experiments were done with 2 biological replicates TNF- α (-) of (**b**), 3 biological replicates TNF- α (+) of (**b**), and 6 biological replicates for (**c**), and data are shown as mean $\pm$ SD. Exact p values ($p < 0.0001$) were reported in the Source Data. Source data are provided as a Source Data file.

Supplementary Table 1: primary and secondary antibodies for IHC.

Reagent or resource	Source	Identifier	Dilution
Primary antibodies for IHC			
Rabbit anti-IBA1	Wako	Cat#: 019-1941	1:500
Rabbit anti-GFAP	Abcam	Cat#: ab53554	1:500
Rabbit anti-RFP	Thermo Fisher Scientific	Cat#: 600-401-379-RTU	1:400
Biotin anti-RFP	Abcam	Cat#: ab34771	1:400
Rabbit anti-CD45	Abcam	Cat#: ab10558	1:200
Rat anti-F4/80	Bio-Rad	Cat#: MCA497G	1:100
Rat anti-B220	Thermo Fisher Scientific	Cat#: 14-0452-82	1:100
Rat anti-NeuN	Millipore	Cat#: MAB377	1:100
Rat anti-CD3	Dako	Cat#: A0452	1:100
Rat anti-Mouse CD8a	BD Pharmingen	Cat#: 553027	1:500
Rabbit anti-GFP	Abcam	Cat#: ab290	1:200
Rabbit anti-Mouse NF-kB p65	Abcam	Cat#: ab16502	1:4000
Secondary antibodies for IHC			
Alexa Flour 488 goat anti-mouse	Thermo Fisher Scientific	Cat#: A11001	1:400
Alexa Fluor 633 goat anti-rabbit	Thermo Fisher Scientific	Cat#: A21070	1:400
Alexa Fluor 568 goat anti-rat	Thermo Fisher Scientific	Cat#: A11077	1:400
Alexa Fluor 568-conjugated streptavidin	Thermo Fisher Scientific	Cat#: S11226	1:400
Alexa Fluor 488 goat anti-rat	Abcam	Cat#: ab150077	1:800

Supplementary Table 2: primary and secondary antibodies for WB.

Reagent or resource	Source	Identifier	Dilution
Primary antibodies for WB			
Rabbit anti-TSG101	Thermo Fisher Scientific	Cat#: PA5-82236	1:1000
Rabbit anti-Cre	Abcam	Cat#: ab188568	1:1000
Rabbit anti-Syntenin-1	Thermo Fisher Scientific	Cat#: 600-401-379-RTU	1:1000
Goat anti-VSV-G	Thermo Fisher Scientific	Cat#: PA1-30278	1:1000
Mouse anti-Cas9	Thermo Fisher Scientific	Cat#: MA5-23519	1:1000
Goat anti-Calnexin	Thermo Fisher Scientific	Cat#: MCA497G	1:1000
Mouse anti- β -Actin	Sigma	Cat#: A5441	1:20000
Mouse anti- I κ B-alpha	Thermo Fisher Scientific	Cat#: MA5-15132	1:1000
Rabbit anti-PCSK9	Thermo Fisher Scientific	Cat#: PA5-78663	1:1000
Secondary antibodies for WB			
IRDye® 800CW Donkey anti-Goat IgG	LI-COR Biosciences	Cat#: 926-32214	1:10000
IRDye® 800CW Goat anti-Mouse IgG	LI-COR Biosciences	Cat#: 926-32210	1:10000
IRDye® 800CW Goat anti-Rabbit IgG	LI-COR Biosciences	Cat#: 926-32211	1:10000
IRDye® 680RD Goat anti-Mouse IgG	LI-COR Biosciences	Cat#: 926-68070	1:10000
IRDye® 800CW Donkey anti-Goat IgG	LI-COR Biosciences	Cat#: 926-32214	1:10000

Supplementary Table 3: plasmids used in this study (1).

Reagent or resource (plasmids)	Source	Identifier
pCMV-VSV-G	Addgene	8454
pLV-CMV-LoxP-DsRed-LoxP-eGFP	Addgene	65726
pCMV-VSV-G-(P127D)-Myc	Addgene	80055
pBIC-Gag-CAS9	Addgene	119942
pBS-CMV-gagpol	Addgene	35614
pLEX-Intein-Cre	This study	
pLEX-CD63-Cre	This study	
pLEX-CD63-Intein-Cre	This study	
pLEX-VSV-G-Intein-Cre	This study	
pLEX-VSV-G-Cre	This study	
pLEX-VSV-G-Foldon-Intein-Cre	This study	
pLEX-VSV-G-Foldon-Intein-MS2	This study	
pLEX-VSV-G-Foldon-Cre	This study	
pLEX-CD63-mnGFP	This study	
pLEX-CD63-Intein-mnGFP	This study	
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pLEX-VSV-G (P127D)-Fodon-Intein-Cre	This study	
pLEX-VSV-G (K47Q)	This study	
pLEX-VSV-G-Foldon-Intein-Cas9	This study	
pLEX-CD63-Intein-Cas9	This study	
pLEX-VSV-G-Intein-SR	This study	
pLEX-VSV-G-Foldon-Intein-SR	This study	
pLEX-super repressor (SR)	This study	
pLEX-CD9-Intein-Cre	This study	
pLEX-C81-Intein-Cre	This study	
pLEX-PTGFRN-Intein-Cre	This study	
pBB-U6-GFPsgRNA	This study	
pLEX-CV-G-Intein-Cre	This study	
pLEX-EPFV-Intein-Cre	This study	
pLEX-CV-G	This study	
pTwist CMV-ERVK-19	This study	
pTwist CMV-ERVMER34-1	This study	
pTwist CMV-ERVI-1	This study	
pTwist CMV-SPACA3	This study	
pTwist CMV-EQTN	This study	
pTwist CMV-TMEM95	This study	
pTwist CMV-HERV-H	This study	
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pTwist CMV-ERVK13-1	This study	
pTwist CMV-FIMP	This study	
pTwist CMV-Syncytin-2	This study	
pTwist CMV-HERVK_113	This study	
pTwist CMV-ERVK-21	This study	
pTwist CMV-Env type 1	This study	
pTwist CMV-SPAM1	This study	
pTwist CMV-ERVK-18	This study	
pTwist CMV-ERVK-8	This study	
pTwist CMV-ERVK-25	This study	
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pTwist CMV-ERVK-9	This study	
pTwist CMV-ERVK-24	This study	
pTwist CMV-Envelope glycoprotein (1)	This study	
pTwist CMV-IZUMO1	This study	
pTwist CMV-HERV-H	This study	

Supplementary Table 4: plasmids used in this study (2).

pTwist CMV-ERVK-7	
pTwist CMV-ERVS71-1	This study
pTwist CMV-Envelope protein	This study
pTwist CMV-Pol/env protein	This study
pTwist CMV-MYMK	This study
pTwist CMV-ERVK-6	This study
pTwist CMV-ERVPALB-1	This study
pTwist CMV-Envelope polyprotein (2)	This study
pTwist CMV-FLJ32214	This study
pTwist CMV-SPACA6	This study
pTwist CMV-ERVFC1	This study
pTwist CMV-ERVV-1	This study
pTwist CMV-Envelope glycoprotein (3)	This study
pTwist CMV-Env type 2	This study
pTwist CMV-MYMX	This study
pTwist CMV-IZUMO1R	This study
pcoPE01(EPFV)	This study
pCD/NL-BH	This study
pLEX-M1 PCSK9	This study
pLEX-CD63-Intein-M1	This study
pLEX-VSV-G-Foldon-Intein-M1	This study
pLEX-CD63-Intein (H439Q)-Cre	This study
pLEX-CD63-Intein (N440A)-Cre	This study
pLEX-VSV-G-Foldon-Intein (H439Q)-Cre	This study
pLEX-VSV-G-Foldon-Intein (N440A)-Cre	This study
pLEX-CD63-Intein-SR	This study

Supplementary Table 5: sgRNA and primer sequences.

Reagent or resource (sequences)	Source	Sequence
sgRNA for HEK-SL	This study	GGACAGTACTCCG CTCGAGT
sgRNA for mTTR	This study	TTACAGCCACGTC TACAGCA
PCR primer for mTTR Forward	This study	gggatcagcatgtactttg gt
Reverse		cttacaagagcaatacgtg ccc

Supplementary Table 6: Reagents and cell lines used in this study.

Reagent or resource	Source	Identifier
Plasmid Plus Midi Kit	QIAGEN	Cat#: 12943
FastScan™ Cas9 (S. pyogenes) ELISA Kit	Cell Signaling Technology	Cat#: 29666
Plasmid Plus Maxi Kit	QIAGEN	Cat#: 12963
Phusion High-Fidelity DNA Polymerase	Thermo Fisher Scientific	Cat#: 15331732
NucleoSpin Gel and PCR Clean-up XS	Tehtum Lab	Cat#: 32-740611.250
HotStarTaq Plus Master Mix Kit	QIAGEN	Cat#: 203645
Maxwell RSC Cell DNA Purification Kit	Promega	Cat#: AS1370
T4 DNA Ligase	Thermo Fisher Scientific	Cat#: EL0011
One Shot TOP10 Chemically Competent E. coli	Thermo Fisher Scientific	Cat#: C404006
Ibidi USA U SLIDE	IBIDI	Cat#: 81806
Millicell Cell Culture Insert	Merck Millipore	Cat#: PICM01250
polyethylenimine (PEI) MAX	Polysciences	Cat#: 24765-1
Intercept™ blocking buffer	LI-COR Biosciences	Cat#: 927-60003
TNF-α	Thermo Fisher Scientific	Cat#: PHC3013

Reagent or resource	Source	Identifier
Experimental cell lines		
HEK-293T	ATCC	Cat#: CRL-3216
HeLa-TL	This study	
Huh7	Elabscience	Cat#: EP-CL-0120
B16F10-TL	This study	
MSC-TL	This study	
Raw264.7-TL	This study	
THP-1-TL	This study	
K562-TL	This study	
HEK-SL	Pieter Vader's lab	
MSC-TL	This study	
T47D-TL	This study	
HEK-NF-κB	InvivoGen	

Supplementary Table 7: Experimental animals used in this study.

Reagent or resource	Source	Identifier
Experimental animals		
C57BL/6J mice	Charles River Laboratories	Cat#: 027
Cre-LoxP R26-LSL-tdTomato reporter mice	Jackson Laboratory	Cat#: 007914

Unprocessed blots of Supplementary figures

Fig. s1d: left panels without VSV-G

 Panels used in the paper

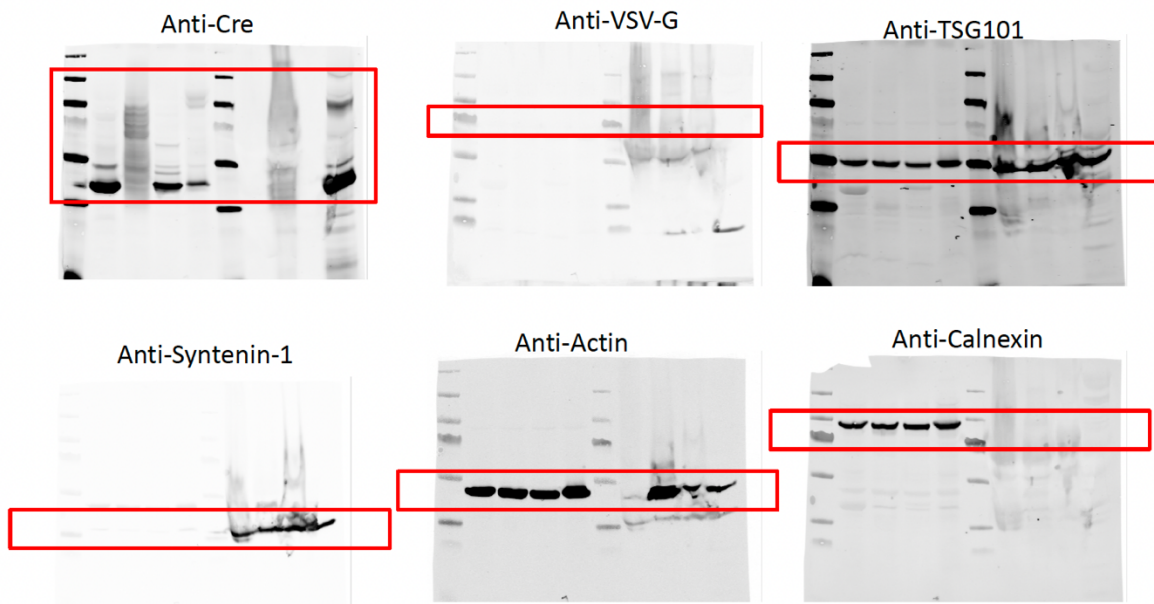


Fig. s1d: right panels with VSV-G

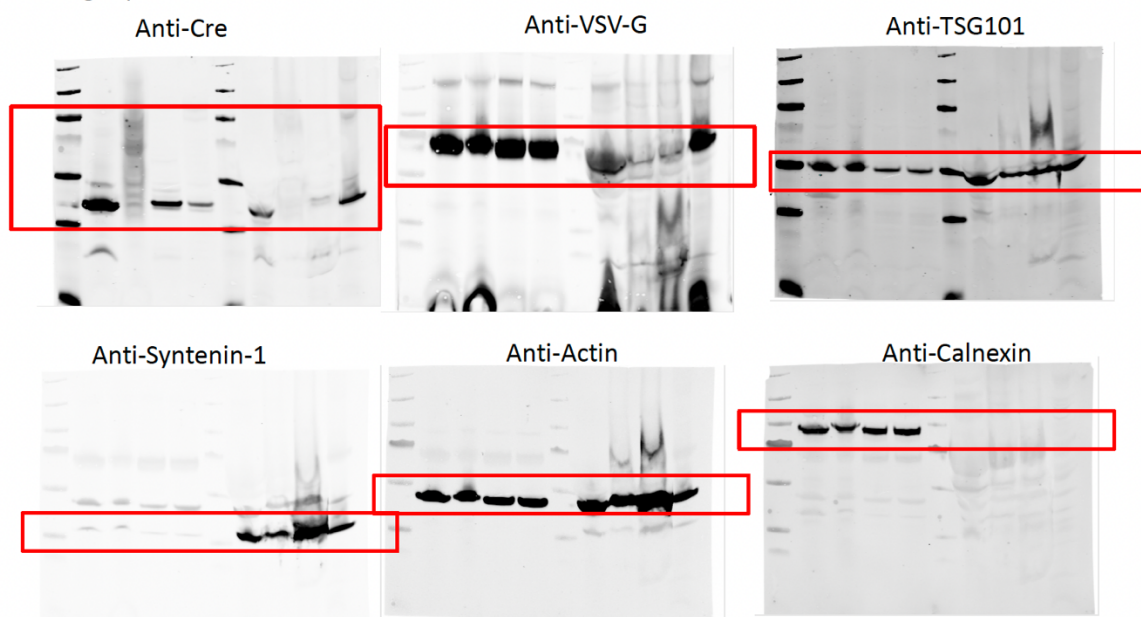


Fig. s2c

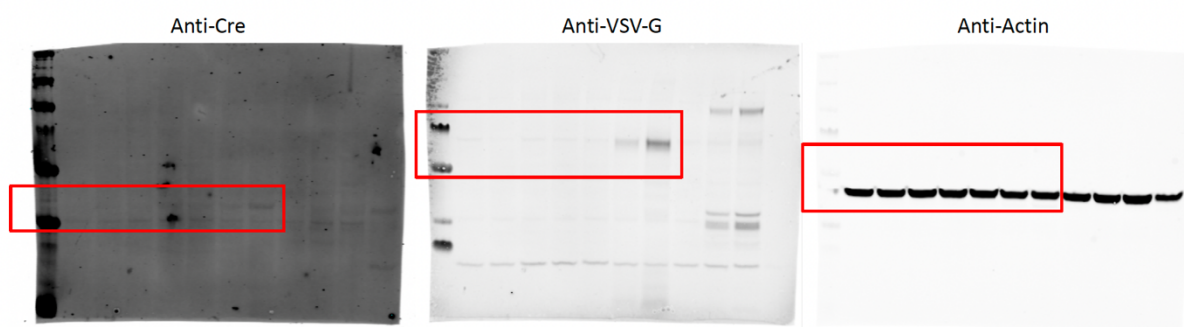


Fig. s4d: left panels

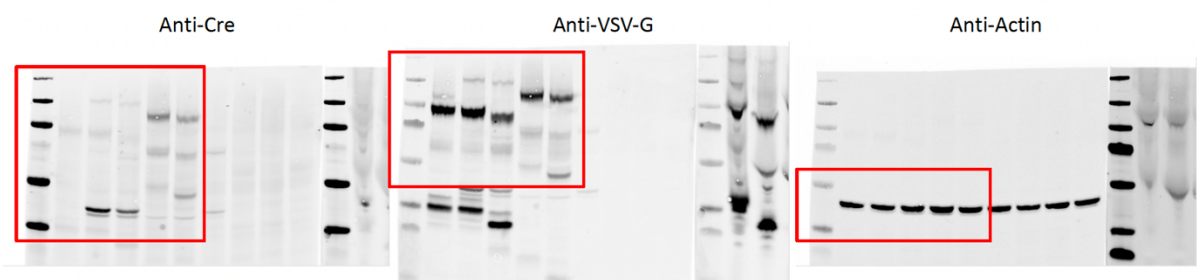


Fig. s4d: right panels

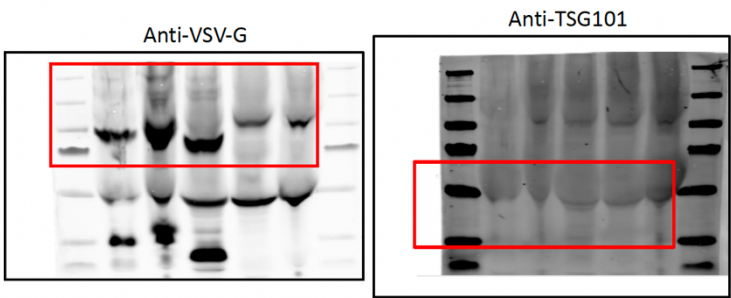


Fig. s6c

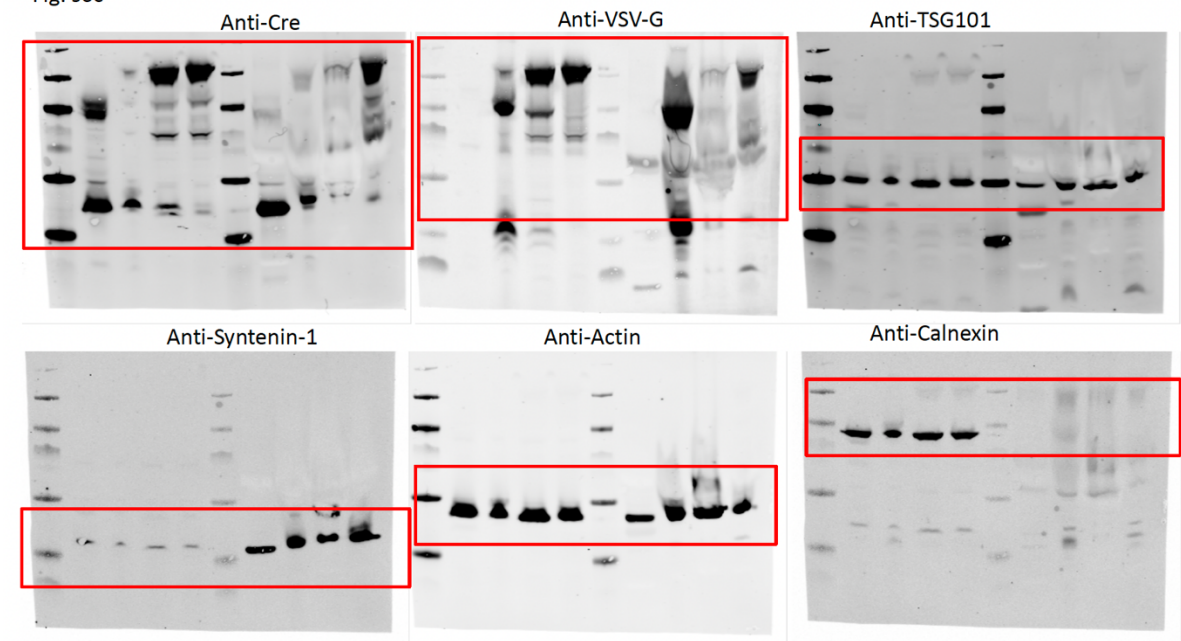


Fig. s9g

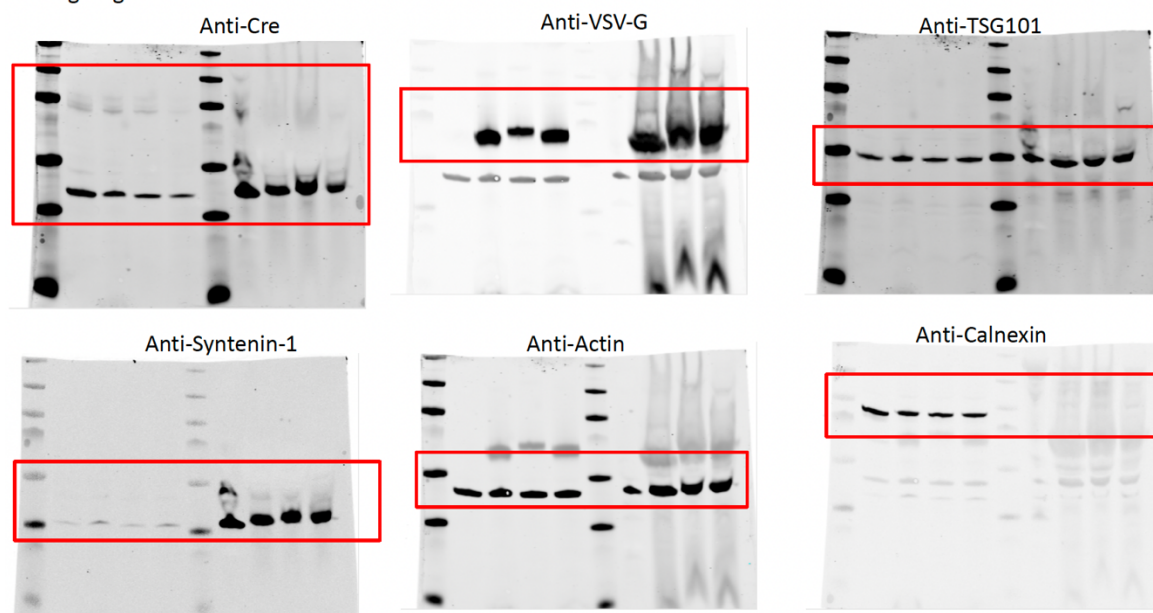


Fig. s10e

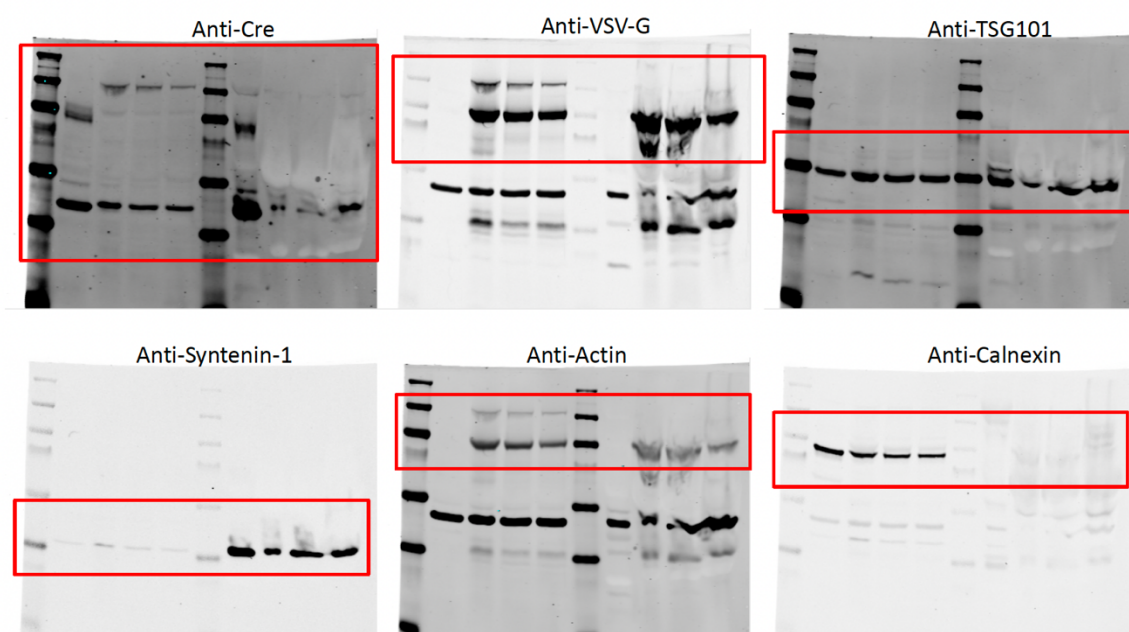


Fig. s11b

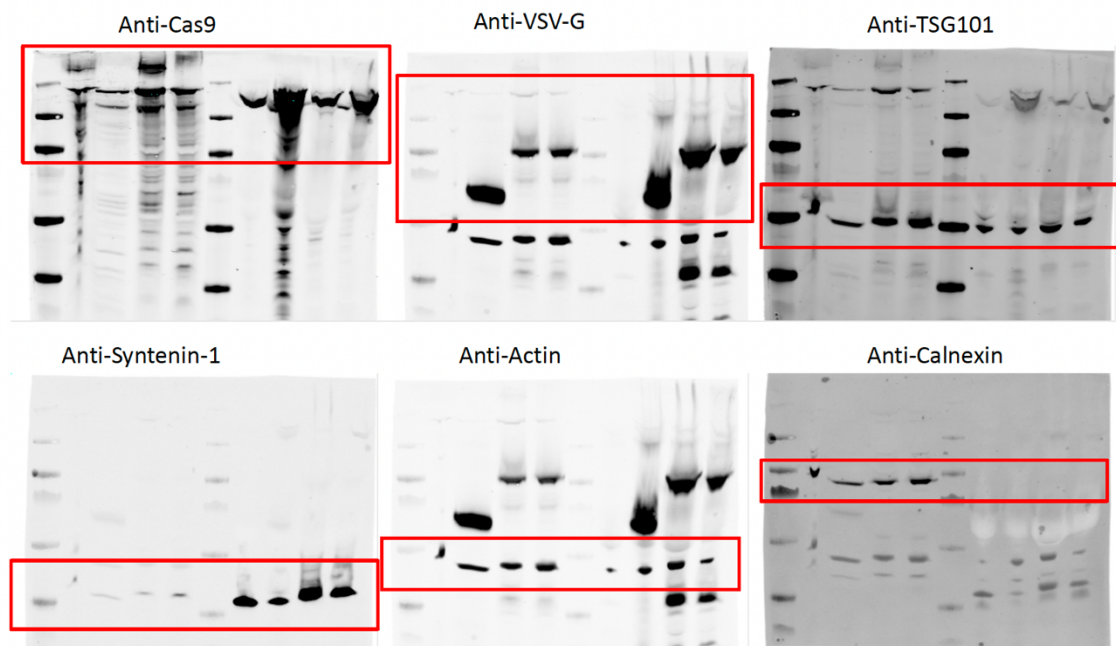


Fig. s11c

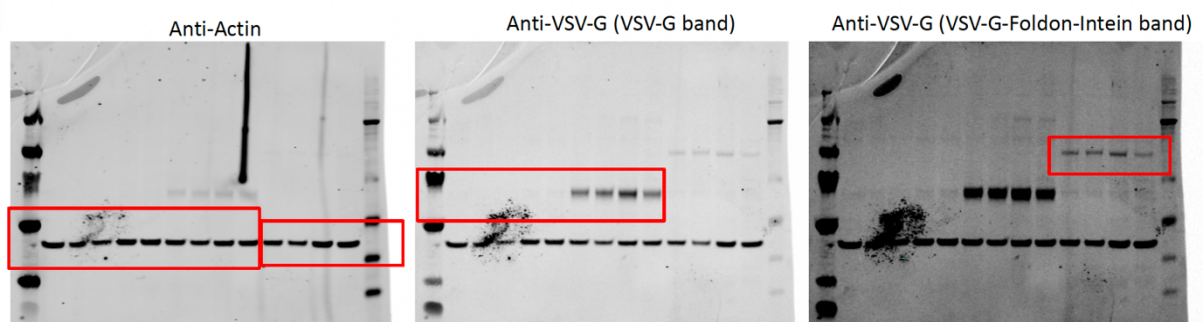


Fig. s16d: left panels

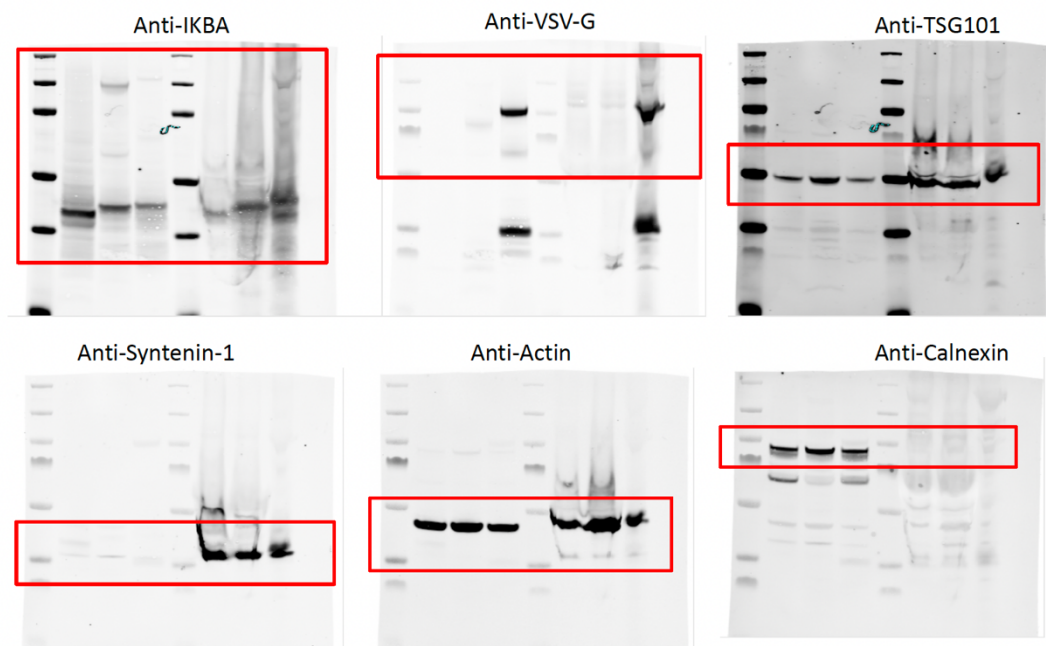


Fig. s16d: right panels

