

Product Sheet

H_IL1RAP IL1R1 KO Reporter HEK-293 Cell Line

Catalog number: GM-C44839

Version 3.3.1.260724

Description	H_IL1RAP IL1R1 KO Reporter HEK-293 Cell Line is a clonal stable cell line derived from 293 cells with a knockout of human IL1RAP and human IL1R1.
Quantity	5E6 Cells per vial, 1 mL
Product Format	1 vial of frozen cells
Shipping	Shipped on dry ice
Storage Conditions	Liquid nitrogen immediately upon receipt
Target	Human IL1RAP & Human IL1R1
Gene ID/Uniprot ID	/
Host Cell	HEK-293
Recovery Medium	DMEM+10% FBS+1% P.S
Growth medium	DMEM+10% FBS+1% P.S+4 µg/mL Blasticidin
Note	None
Freezing Medium	90% FBS+10% DMSO
Growth properties	Adherent
Growth Conditions	37°C, 5% CO ₂
Mycoplasma Testing	The cell line has been screened to confirm the absence of Mycoplasma species.
Safety considerations	Biosafety Level 2
Note	It is recommended to expand the cell culture and store a minimum of 10 vials at an early passage for potential future use.

Materials

Reagent	Manufacturer/Catalogue No.
DMEM	Gibco/C11995500BT
Fetal Bovine Serum	ExCell/FSP500
Pen/Strep	Thermo/15140-122
Blasticidin	Genomeditech/GM-040404
Human IL-1 alpha Protein; His Tag	Genomeditech/GM-88245RP
Human IL-1 beta Protein; His Tag	Genomeditech/GM-88248RP
Anti-IL1RAP hIgG1 Antibody (48D2_VH5.GL_VL4)	Genomeditech/GM-88388AB

Figures

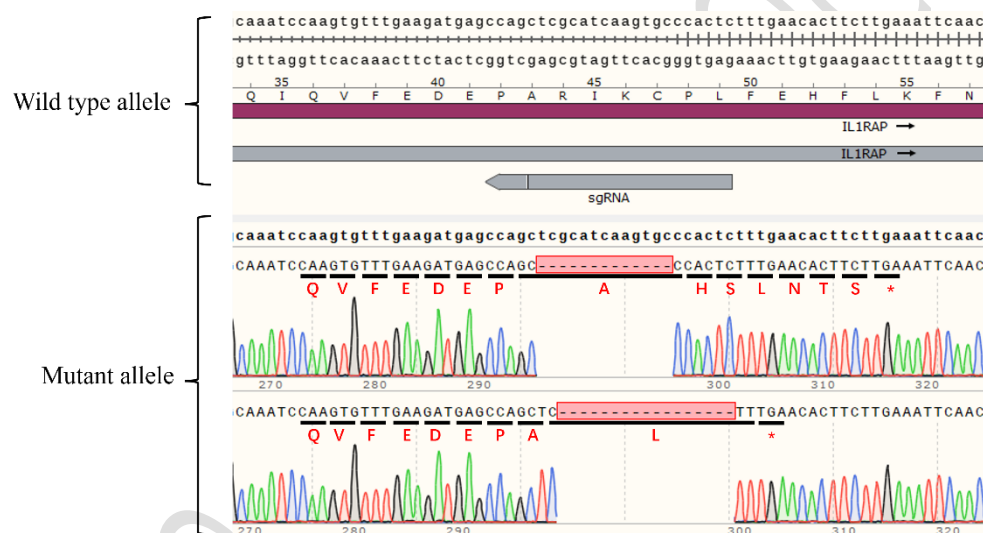


Figure 1 | The Sanger sequencing of the H_IL1RAP IL1R1 KO Reporter HEK-293 Cell Line(Cat. GM-C44839) showed successful knockout of IL1RAP.



Figure 2 | The Sanger sequencing of the H_IL1RAP IL1R1 KO Reporter HEK-293 Cell Line(Cat. GM-C44839) showed successful knockout of IL1R1.

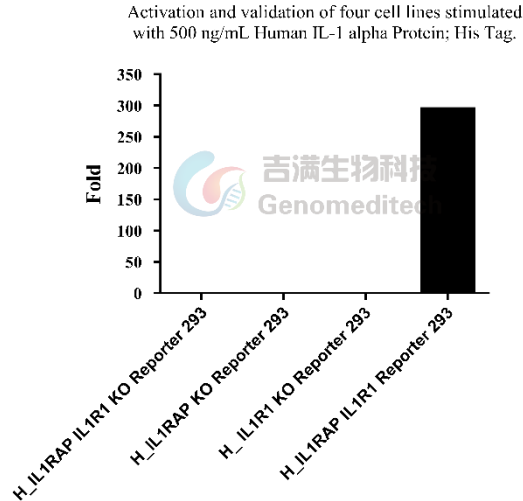


Figure 3 | Response to IL-1 alpha Protein. The H_IL1RAP IL1R1 KO Reporter 293 (Cat. GM-C44839)(IL1RAP/IL1R1 double KO), H_IL1RAP KO Reporter 293(double KO + IL1R1 rescue), H_IL1R1 KO Reporter 293(double KO + IL1RAP rescue), and H_IL1RAP IL1R1 Reporter 293(double KO + IL1RAP/IL1R1 rescue) at a concentration of 1.5×10^4 cells/well (96-well format) were stimulated with 500 ng/mL Human IL-1 alpha Protein (Cat. [GM-88245RP](#)) in assay buffer for 6 hours. The firefly luciferase activity was measured using the Luciferase Reporter Assay Kit. Only the cell line co-expressing both IL1RAP and IL1R1 (the H_IL1RAP IL1R1 Reporter) exhibited significant activation, whereas the single-reconstituted lines (IL1RAP KO or IL1R1 KO) showed no response, confirming that intact IL-1 signaling requires both receptor components.

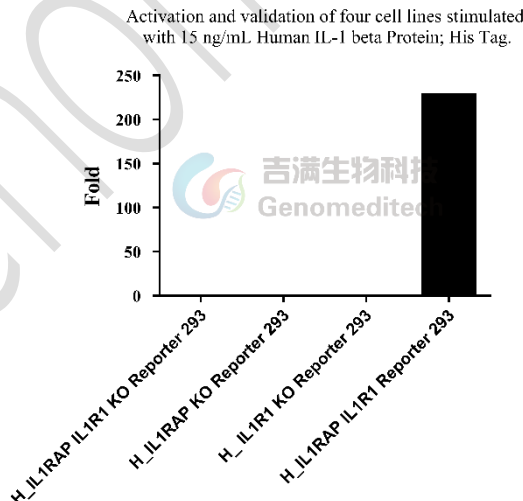


Figure 4 | Response to IL-1 beta Protein. The H_IL1RAP IL1R1 KO Reporter 293 (Cat. GM-C44839)(IL1RAP/IL1R1 double KO), H_IL1RAP KO Reporter 293(double KO + IL1R1 rescue), H_IL1R1 KO Reporter 293(double KO + IL1RAP rescue), and H_IL1RAP IL1R1 Reporter 293(double KO + IL1RAP/IL1R1 rescue) at a concentration of 1.5×10^4 cells/well (96-well format) were stimulated with 15 ng/mL Human IL-1 beta Protein (Cat. [GM-88248RP](#)) in assay buffer for 6 hours. The firefly luciferase activity was measured using the Luciferase Reporter Assay Kit. Only the cell

line co-expressing both IL1RAP and IL1R1 (the H_IL1RAP IL1R1 Reporter) exhibited significant activation, whereas the single-reconstituted lines (IL1RAP KO or IL1R1 KO) showed no response, confirming that intact IL-1 signaling requires both receptor components.

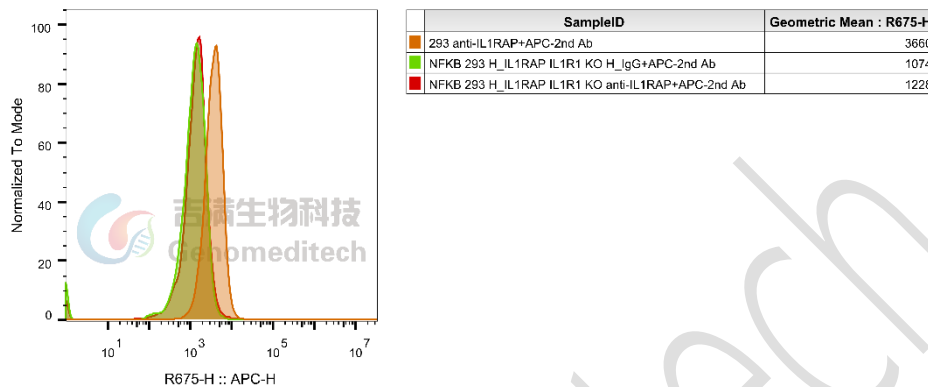


Figure 5 | H_IL1RAP IL1R1 KO Reporter HEK-293 Cell Line (Cat. GM-C44839) was determined by flow cytometry using Anti-IL1RAP hIgG1 Antibody (48D2_VH5.GL_VL4) (Cat. GM-88388AB).

Cell Recovery

Recovery Medium: DMEM+10% FBS+1% P.S

To insure the highest level of viability, thaw the vial and initiate the culture as soon as possible upon receipt. If upon arrival, continued storage of the frozen culture is necessary, it should be stored in liquid nitrogen vapor phase and not at -70°C. Storage at -70°C will result in loss of viability.

- Thaw the vial by gentle agitation in a 37°C water bath. To reduce the possibility of contamination, keep the O-ring and cap out of the water. Thawing should be rapid (approximately 2 - 3 minutes).
- Remove the vial from the water bath as soon as the contents are thawed, and decontaminate by dipping in or spraying with 70% ethanol. All of the operations from this point on should be carried out under strict aseptic conditions.
- Transfer the vial contents to a centrifuge tube containing 5.0 mL complete culture medium. And spin at approximately 176 x g for 5 minutes. Discard supernatant.
- Resuspend cell pellet with the recommended complete medium. And dispense the suspension into an appropriate culture flask and initially place the flask in an upright position after thawing.
- Incubate the culture at 37°C in a suitable incubator. A 5% CO₂ in air atmosphere is recommended if using the medium described on this product sheet.

Cell Freezing

Freezing Medium: 90% FBS+10% DMSO

- Centrifuge at 176 x g for 3 minutes to collect cells.
- Resuspend the cells in pre-cooled freezing medium and adjust the cell density to 5E6 cells/mL.
- Aliquot 1 mL into each vial.

- d) Place the vial in a controlled-rate freezing container and store at -80°C for at least 1 day, then transfer to liquid nitrogen as soon as possible.

Cell passage

Growth medium: DMEM+10% FBS+1% P.S+4 µg/mL Blasticidin

For the first 1 to 2 passages post-resuscitation, use the recovery medium. Once the cells have stabilized, switch to a growth medium.

- Remove and discard culture medium.
- Briefly rinse the cell layer with PBS to remove all traces of serum that contains trypsin inhibitor.
- Add 1.0 mL of 0.25% (w/v) Trypsin-EDTA solution to dish and observe cells under an inverted microscope until cell layer is dispersed (usually within 30 to 60 seconds at 37°C).
- Note: To avoid clumping do not agitate the cells by hitting or shaking the flask while waiting for the cells to detach. Cells that are difficult to detach may be placed at 37°C to facilitate dispersal.
- Add 2.0 mL of growth medium to mix well and aspirate cells by gently pipetting.
- After centrifugation, resuspend the pellet and add appropriate aliquots of the cell suspension to new culture vessels.
- Incubate cultures at 37°C.

Subcultivation Ratio: A subcultivation ratio of 1:3 - 1:4 is recommended

Medium Renewal: Every 3 days

Notes

- After initial thawing, a higher proportion of dead cells is normal. The cell culture generally improves noticeably after approximately one week of adaptation. Once the culture stabilizes, the percentage of dead cells decreases with subsequent passages, and the cell proliferation rate becomes more consistent.
- It is important to maintain the cell density below 80%, as exceeding this threshold can lead to decreased cell viability and metabolic activity due to overcrowding.
- FBS requires heat inactivation at 56°C for 30 minutes, which can inactivate complement and some viruses, but does not significantly affect the activity of most growth factors and cytokines.

Related Products

IL-1	
H_IL1RAP KO HEK-293 Cell Line	H_IL-1 Reporter 293 Cell Line
H_IL-1 Reporter 293 Cell Line (old version)	Cynomolgus_IL-1R HEK-293 Cell Line
Cynomolgus_IL1RAP HEK-293 Cell Line	H_IL-1R CHO-K1 Cell Line
H_IL-1R HEK-293 Cell Line	H_IL1RAP CHO-K1 Cell Line
H_IL1RAP HEK-293 Cell Line	Membrane bound H_IL-1α CHO-K1 Cell Line
Membrane bound H_IL-1α HEK-293 Cell Line	Membrane bound H_IL-1β CHO-K1 Cell Line
Membrane bound H_IL-1β HEK-293 Cell Line	Mouse_IL1RAP HEK-293 Cell Line
Rat_IL1RAP HEK-293 Cell Line	

Anti-IL1B×IL1A hIgG1 Bispecific antibody(lutikizumab)	Anti-IL1R1 hIgG1 Antibody(AMG 108)
Anti-IL1RAP hIgG1 Antibody (Nadunolimab)	Anti-IL-1β hIgG1 Antibody(Canakinumab)
Biotinylated Human IL-1 alpha Protein; His-Avi Tag	Biotinylated Human IL-1 beta Protein; His-Avi Tag
Cynomolgus IL-1 alpha Protein; His Tag	Cynomolgus IL-1 beta Protein; His Tag
Human IL-1 alpha Protein; hFc Tag	Human IL-1 alpha Protein; His Tag
Human IL-1 beta Protein; hFc Tag	Human IL-1 beta Protein; His Tag
Human IL-1RAP Protein; hFc Tag	Human IL-1RAP Protein; His Tag
Human IL-1RI Protein; hFc Tag	Human IL-1RI Protein; His Tag
Human IL-1RI&IL-1RAP Heterodimer Protein; hFc Tag (Goflikicept)	

License Agreement:

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