

# Product Sheet

## H\_CD40(TNFRSF5) Reporter 293 Cell Line

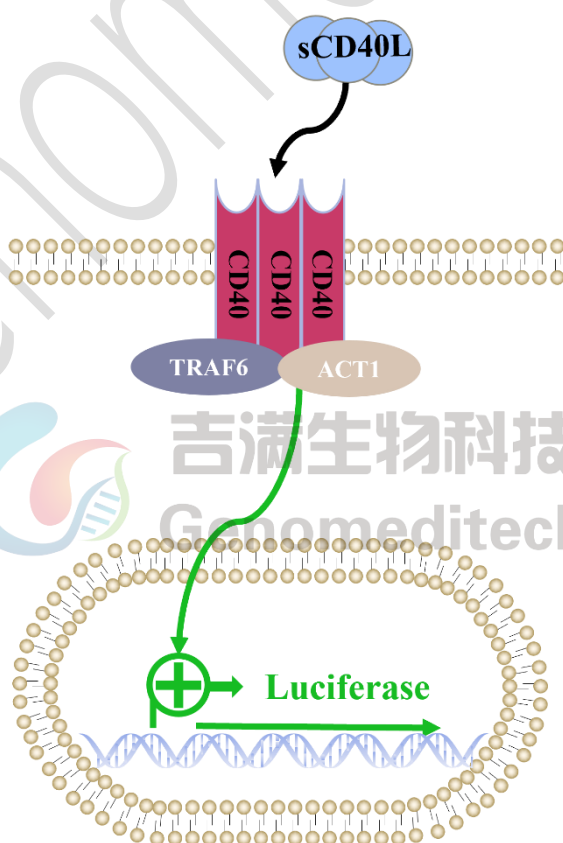
Catalog number: GM-C03829

Version 3.3.1.251219

CD40 is a key cell surface receptor in the TNF receptor superfamily, primarily found on B cells, dendritic cells, macrophages, and some endothelial cells. Its main role is to regulate immune responses by promoting B cell proliferation, differentiation, and antibody production. When it binds to its ligand CD40L (expressed by activated T cells), CD40 activates various signaling pathways that enhance immune function.

CD40 signaling occurs through multiple pathways activated upon CD40L binding, including NF- $\kappa$ B, MAPK (ERK, JNK, p38 MAPK), and PI3K/Akt. This activation triggers biological responses such as cell proliferation, survival, cytokine secretion, and immune memory formation, making CD40 crucial for regulating immune responses and maintaining immune tolerance.

H\_CD40(TNFRSF5) Reporter 293 Cell Line is a clonal stable HEK-293 cell line constructed using lentiviral technology, constitutive expression of the CD40 gene, along with signal-dependent expression of a luciferase reporter gene. When CD40L binds to CD40, it activates downstream signaling pathways, leading to the expression of luciferase. The luciferase activity measurement indicates the activation level of the signaling pathway and can thus be used to evaluate the in vitro effects of drugs related to CD40.



## Specifications

<b>Quantity</b>	5E6 Cells per vial, 1 mL
<b>Product Format</b>	1 vial of frozen cells
<b>Shipping</b>	Shipped on dry ice
<b>Storage Conditions</b>	Liquid nitrogen immediately upon receipt

<b>Recovery Medium</b>	DMEM+10% FBS+1% P.S
<b>Growth medium</b>	EMEM+10% FBS+1% P.S+3 µg/mL Blasticidin+1.5 µg/mL Puromycin
<b>Note</b>	None
<b>Freezing Medium</b>	90% FBS+10% DMSO
<b>Growth properties</b>	Adherent
<b>Growth Conditions</b>	37°C, 5% CO <sub>2</sub>

<b>Mycoplasma Testing</b>	The cell line has been screened to confirm the absence of Mycoplasma species.
<b>Safety considerations</b>	Biosafety Level 2
<b>Note</b>	It is recommended to expand the cell culture and store a minimum of 10 vials at an early passage for potential future use.

## Materials

<b>Reagent</b>	<b>Manufacturer/Catalogue No.</b>
EMEM	ATCC/30-2003
Fetal Bovine Serum	ExCell/FSP500
Pen/Strep	Thermo/15140-122
Blasticidin	Genomeditech/ <a href="#">GM-040404</a>
Puromycin	Genomeditech/ <a href="#">GM-040401</a>
Human CD40L Protein; His Tag	Genomeditech/ <a href="#">GM-84919RP</a>
Anti-CD40 hIgG1 Reference Antibody (Tenebio)	Genomeditech/GM-87983MAB
GMOne-Step 2.0 Luciferase Reporter Gene Assay Kit	Genomeditech/ <a href="#">GM-040513</a>

## Figures

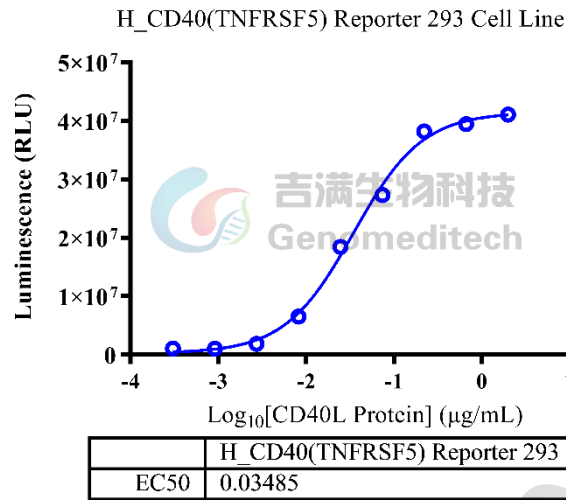


Figure 1 | Response to Human CD40L Protein; His Tag. The H\_CD40(TNFRSF5) Reporter 293 Cell Line (Cat. GM-C03829) at a concentration of 1.5E4 cells/well (96-well format) was stimulated with serial dilutions of Human CD40L Protein (Cat. [GM-84919RP](#)) in assay buffer (DMEM+1% FBS+1% P.S) for 6 hours. The firefly luciferase activity was measured using the Luciferase Reporter Assay Kit (Genomeditech). The maximum induction fold was approximately [44.2]. Data are shown by drug mass concentration.

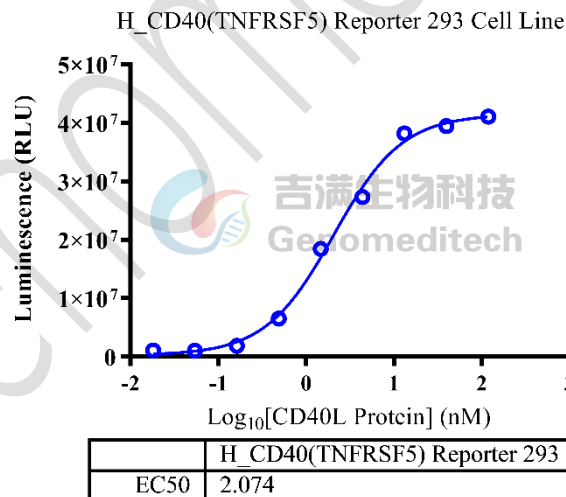


Figure 2 | Response to Human CD40L Protein; His Tag. The H\_CD40(TNFRSF5) Reporter 293 Cell Line (Cat. GM-C03829) at a concentration of 1.5E4 cells/well (96-well format) was stimulated with serial dilutions of Human CD40L Protein (Cat. [GM-84919RP](#)) in assay buffer (DMEM+1% FBS+1% P.S) for 6 hours. The firefly luciferase activity was measured using the Luciferase Reporter Assay Kit (Genomeditech). The maximum induction fold was approximately [44.2]. Data are shown by drug molar concentration.

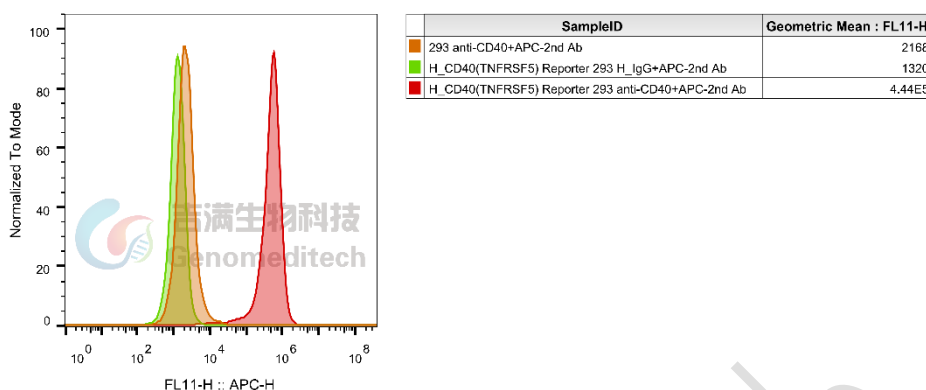


Figure 3 | H\_CD40(TNFRSF5) Reporter 293 Cell Line(Cat. GM-C03829) was determined by flow cytometry using Anti-CD40 hIgG1 Reference Antibody (Tenebio) (Cat. GM-87983MAB).

## 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 recovery medium. And dispense into appropriate culture dishes.
- Incubate the culture at 37°C in a suitable incubator. A 5% CO<sub>2</sub> 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.
- 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: EMEM+10% FBS+1% P.S+3 µg/mL Blasticidin+1.5 µg/mL Puromycin

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 2 to 3 days**

## Notes

- Upon initial revival, a higher number of dead cells and poor adherence are observed, which is normal. Adherence typically recovers within 2 - 3 days. After 2 - 3 passages, the proportion of adherent cells increases, and the cells begin to spread normally.
- After each passage, there may be 5 - 10% dead cells; however, as the number of passages increases, the recovery rate accelerates, the proportion of dead cells decreases, and the cell growth rate stabilizes.
- It is recommended to retain cell images after revival and during each observation to assist in assessing cell status. In case of abnormalities, promptly communicate with Genomeditech sales.

## Related Products

CD40:CD40L	
<a href="#">H_CD40(TNFRSF5) Reporter Jurkat Cell Line</a>	<a href="#">Cynomolgus_CD40 CHO-K1 Cell Line</a>
<a href="#">Cynomolgus_CD40L CHO-K1 Cell Line</a>	<a href="#">H_CD40(TNFRSF5) CHO-K1 Cell Line</a>
<a href="#">H_CD40(TNFRSF5) HEK-293 Cell Line</a>	<a href="#">H_CD40L CHO-K1 Cell Line</a>
<a href="#">H_CD40L HEK-293 Cell Line</a>	<a href="#">Rabbit_CD40L NIH-3T3 Cell Line</a>
<a href="#">Anti-CD40L hIgG1 Reference Antibody (Frebio)</a>	<a href="#">Anti-H_CD40 hIgG1 Antibody(APX005M)</a>
<a href="#">Anti-H_CD40 hIgG1 Antibody(ravagalimab)</a>	<a href="#">Anti-H_CD40L hIgG1 Antibody(dapirolizumab)</a>
<a href="#">Anti-H_CD40L hIgG1 Antibody(frexalimab)</a>	
<a href="#">Biotinylated Human CD40 Protein; His-Avi Tag</a>	<a href="#">Cynomolgus CD40 Protein; His Tag</a>
<a href="#">Human CD40 Protein; His Tag</a>	<a href="#">Human CD40L Protein; His Tag</a>
IFN-α	

IFN $\alpha$ Reporter HEK-293 Cell Line	IFN $\alpha$ Reporter MDCK Cell Line
IFN $\alpha$ Reporter THP1 Cell Line	
BCMA:BAFFR:TACI	
H_BAFFR Jurkat Blockade Reporter Cell Line	H_BAFFR Reporter Cell Line
H_BCMA Reporter Cell Line	H_TACI Reporter Cell Line
Cynomolgus_BCMA CHO-K1 Cell Line	Cynomolgus_BCMA HEK-293 Cell Line
H_BCMA CHO-K1 Cell Line	H_BCMA HEK-293 Cell Line
Anti-BAFF hIgG1 Antibody(belimumab)	Anti-BAFFR hIgG1 Antibody(inalumab)
Anti-BCMA hIgG1 Antibody(Belantamab)	Anti-BCMA hIgG1 Antibody(SEA-BCMA)
Anti-BCMA hIgG4 Antibody(BCMB69)	
Biotinylated Human BAFF Protein; His-Avi Tag	Biotinylated Human BCMA Protein; His-Avi Tag
Cynomolgus BAFF Protein; His Tag	Cynomolgus BCMA Protein; hFc Tag
Cynomolgus BCMA Protein; His Tag	Human APRIL Protein; hFc Tag
Human BAFF Protein; His Tag	Human BCMA Protein; hFc Tag
Human BCMA Protein; His Tag	Mouse BAFF Protein; His Tag
BDCA2(CLEC4C)	
H_BDCA2 Reporter DDX35TM Jurkat Cell Line	H_BDCA2 Reporter Jurkat Cell Line
Cynomolgus_BDCA2 CHO-K1 Cell Line	Cynomolgus_BDCA2 Jurkat Cell Line
H_BDCA2 CHO-K1 Cell Line	H_BDCA2 HEK-293 Cell Line
H_BDCA2 Jurkat Cell Line	
Anti-H_BDCA2 hIgG1 Antibody(Litifilimab)	
Cynomolgus BDCA2 Protein; His Tag	Human BDCA2 Protein; His Tag

## License Agreement:

**By purchasing and using this cell line product, the user voluntarily agrees to accept and abide by the following policies:**

- This cell line product is restricted to research use only and shall not be used for any commercial purposes.
- This product is strictly prohibited from being used in the diagnosis or treatment of human or animal diseases, and shall not be directly used in experiments involving humans.
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