Genomeditech (Shanghai) Co.,Ltd.
Order: +86 021-68455258/50432826/50432825
Toll-free: +86 400 627 9288
Email: service@genomeditech.com

Product Sheet

H_BCMA Reporter 293 Cell Line

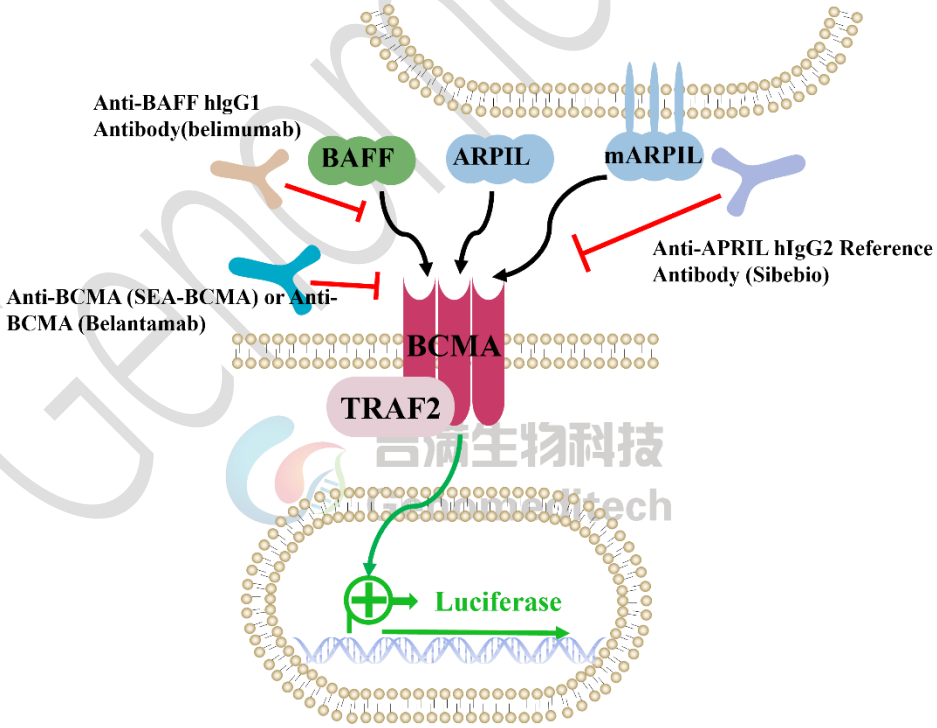
Catalog number: GM-C44705

Version 3.3.1.260724

BCMA (B-cell maturation antigen) is a protein expressed on mature B cells and plasma cells, regulating their survival, proliferation, and differentiation. It is crucial in B cell-related diseases, particularly multiple myeloma (MM), where its levels are elevated, making it a key therapeutic target.

The BCMA signaling pathway is mainly activated by binding to its ligands (such as APRIL and BAFF), which triggers downstream signal transduction. The activation of BCMA can promote the survival and proliferation of B cells through signaling pathways such as NF-κB, MAPK, and PI3K/Akt. The activation of these signaling pathways not only aids in the development and function of B cells but is also closely related to the survival and drug resistance of tumor cells.

H_BCMA Reporter 293 Cell Line is a clonal stable 293 cell line constructed using lentiviral technology, constitutive expression of the BCMA gene, along with signal-dependent expression of a luciferase reporter gene. When Ligand binds to BCMA, it activates downstream signaling pathways, leading to the expression of luciferase. Blockade antibodies can inhibit this signal transmission. 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 BCMA.



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Specifications

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
Recovery Medium	DMEM+10% FBS+1% P.S
Growth medium	DMEM+10% FBS+1% P.S+4 μg/mL Blasticidin+0.75 μg/mL Puromycin
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
Puromycin	Genomeditech/GM-040401
Membrane Bound H_APRIL(Trimer) HEK-293 Cell Line	Genomeditech/GM-C40503
Human BAFF Protein; His Tag	Genomeditech/GM-87735RP
Recombinant Human APRIL (N-Flag-His)	Novoprotein/CU89
Anti-BCMA hIgG1 Antibody(Belantamab)	Genomeditech/GM-52396AB
Anti-TNFSF13B(BAFF) hIgG1 Reference Antibody (Belibio)	Genomeditech/GM-87985MAB
Anti-APRIL hIgG2 Reference Antibody (Sibebio)	Genomeditech/GM-88014MAB
Anti-BCMA hIgG1 Antibody(SEA-BCMA)	Genomeditech/GM-49486AB
GMOne-Step 2.0 Luciferase Reporter Gene Assay Kit	Genomeditech/GM-040513

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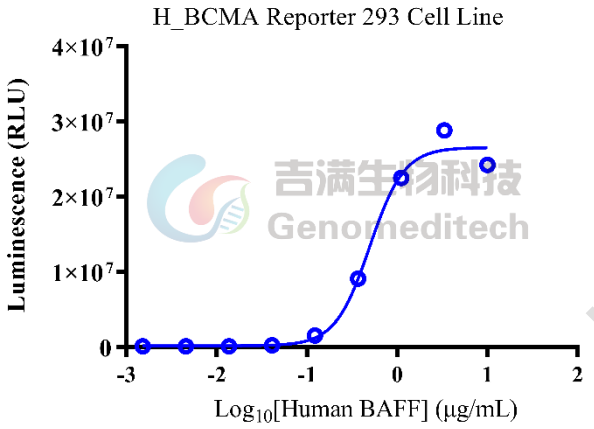
Figures



	H_BCMA Reporter 293 Cell Line
EC50	0.4976

Figure 1 | Response to Human BAFF Protein; His Tag. The H_BCMA Reporter 293 Cell Line (Cat. GM-C44705) at a density of 1.5E4 cells/well (96-well format) was stimulated with serial dilutions of Human BAFF Protein; His Tag(Cat. GM-87735RP) 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 [299.5]. Data are shown by drug mass concentration.

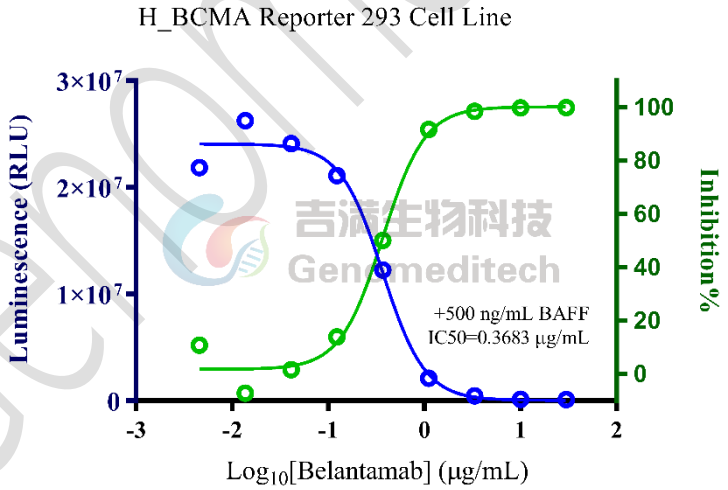


Figure 2 | Inhibition of BAFF Protein-induced reporter activity by Belantamab. H_BCMA Reporter 293 Cell Line (Cat. GM-C44705) was seeded at a density of 1.5E4 cells per well in a 96-well plate and incubated overnight. The next day, Serial dilutions of the Anti-BCMA hIgG1 Antibody(Belantamab)(Cat. GM-52396AB) was incubated with 1.5E4 cells/well of the H_BCMA Reporter 293 Cell Line in a 96-well plate for 1 hour in assay buffer. Subsequently, the Human BAFF Protein (Cat. GM-87735RP) at a density of 50 ng/well was added, and the coculture proceeded for an additional 6 hours. Firefly luciferase activity was then measured using the Luciferase Reporter Assay Kit (Genomeditech). (left Y-axis, relative luminescence units), with inhibition percentages shown on the right Y-axis.

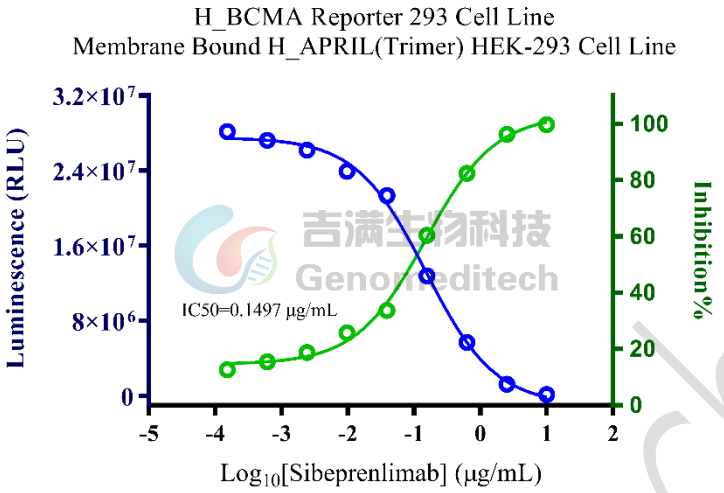


Figure 3 | Inhibition of Membrane Bound H_APRIL(Trimer) 293-induced reporter activity by Sibeprenlimab. The H_BCMA Reporter 293 Cell Line (Cat. GM-C44705) was seeded at a density of 1.5E4 cells per well in a 96-well plate and incubated overnight. The next day, Serial dilutions of the Anti-APRIL hIgG2 Reference Antibody (Sibebio) (Cat. GM-88014MAB) and 1E4 cells/well of the Membrane Bound H_APRIL(Trimer) HEK-293 Cell Line (Cat. GM-C40503) were added to 1.5E4 cells/well of H_BCMA Reporter 293 Cell Line(Cat. GM-C44705) for 6 hours. The firefly luciferase activity was measured using the Luciferase Reporter Assay Kit (Genomeditech). (left Y-axis, relative luminescence units), with inhibition percentages shown on the right Y-axis. Data are shown by drug mass concentration.

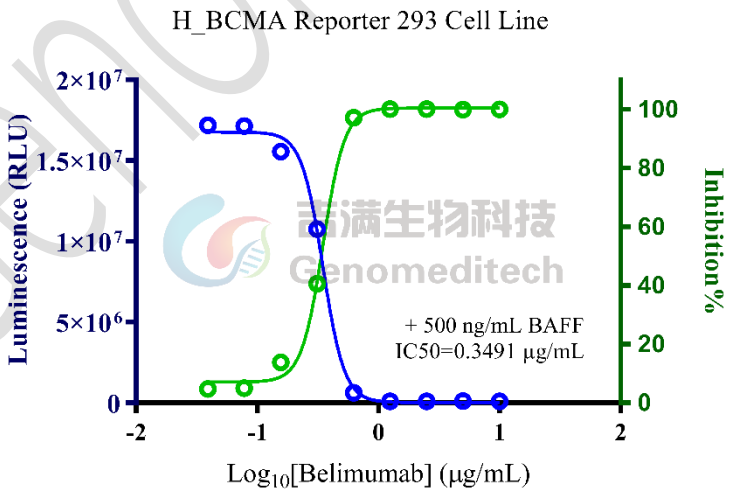
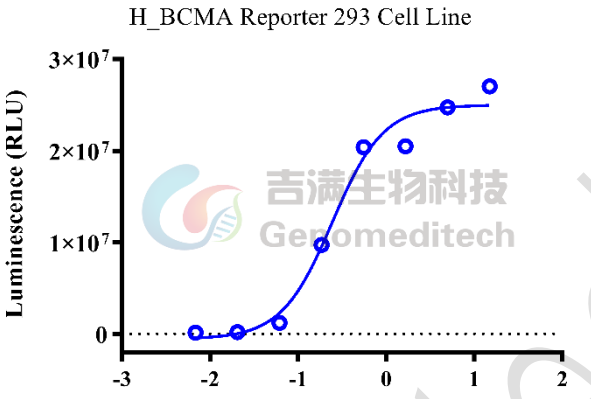


Figure 4 | Inhibition of BAFF Protein-induced reporter activity by Belimumab.H_BCMA Reporter 293 Cell Line (Cat. GM-C44705) was seeded at a density of 1.5E4 cells per well in a 96-well plate and incubated overnight. The next day, Serial dilutions of Anti-TNFSF13B(BAFF) hIgG1 Reference Antibody (Belibio) (Cat. GM-87985MAB) was incubated with 50 ng/mL of Human BAFF Protein(Cat. GM-87735RP) for 1 hour in assay buffer. After pre-incubation,

add to the pre-seeded cells, and incubate for 6 hours. Firefly luciferase activity was then measured using the Luciferase Reporter Assay Kit (Genomeditech). (left Y-axis, relative luminescence units), with inhibition percentages shown on the right Y-axis. Data are shown by drug mass concentration.



	H_BCMA Reporter 293 Cell Line
EC50	0.2459

Figure 5 | Response to Recombinant Human APRIL (N-Flag-His). The H_BCMA Reporter 293 Cell Line (Cat. GM-C44705) at a density of 1.5E4 cells/well (96-well format) was stimulated with serial dilutions of Recombinant Human APRIL (N-Flag-His)(Novoprotein/CU89) 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 [180.2]. Data are shown by drug mass concentration.

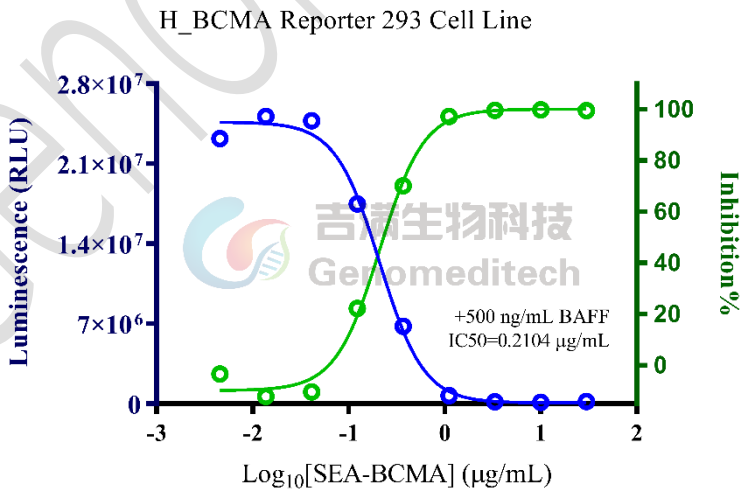


Figure 6 | Inhibition of BAFF Protein-induced reporter activity by SEA-BCMA. H_BCMA Reporter 293 Cell Line (Cat. GM-C44705) was seeded at a density of 1.5E4 cells per well in a 96-well plate and incubated overnight. The next day, Serial dilutions of the Anti-BCMA hIgG1 Antibody(SEA-BCMA)(Cat. GM-49486AB) was incubated with 1.5E4 cells/well of the H_BCMA Reporter 293 Cell Line in a 96-well plate for 1 hour in assay buffer. Subsequently, the

Human BAFF Protein (Cat. GM-87735RP) at a density of 50 ng/well was added, and the coculture proceeded for an additional 6 hours. Firefly luciferase activity was then measured using the Luciferase Reporter Assay Kit (Genomeditech). (left Y-axis, relative luminescence units), with inhibition percentages shown on the right Y-axis.

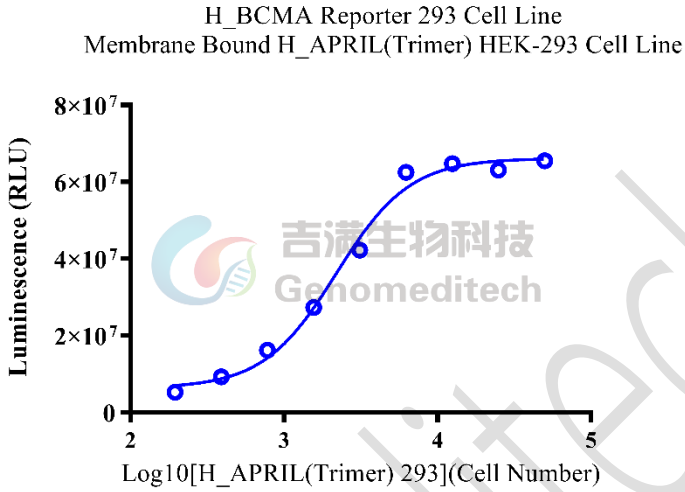


Figure 7 | Response to Membrane Bound H_APRIL(Trimer) HEK-293 Cell Line. H_BCMA Reporter 293 Cell Line (Cat. GM-C44705) at a concentration of 1.5E4 cells/well (96-well format) was stimulated with serial dilutions of Membrane Bound H_APRIL(Trimer) HEK-293 Cell Line (Cat. GM-C40503) for 6 hours. The firefly luciferase activity was measured using the Luciferase Reporter Assay Kit (Genomeditech).

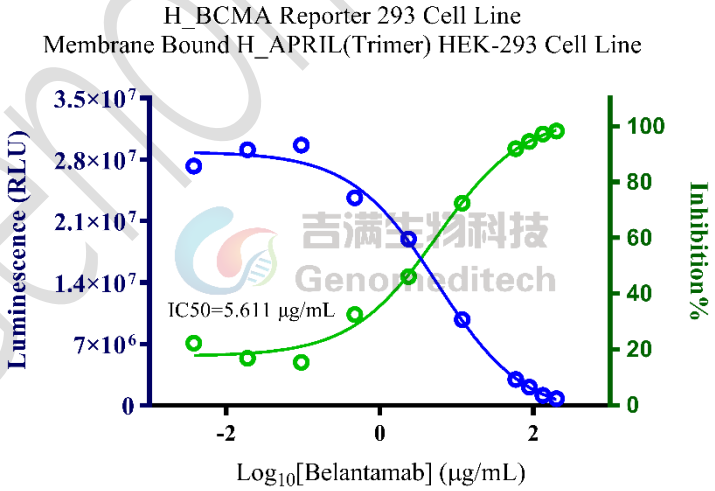


Figure 8 | Belantamab inhibits membrane-bound H_APRIL(Trimer)-induced reporter activity in H_BCMA Reporter 293 cells. The H_BCMA Reporter 293 (Cat. GM-C44705) was seeded at a density of 1.5E4 cells per well in a 96-well plate and incubated overnight. The next day, H_BCMA Reporter 293 Cells were incubated with serial dilutions of Anti-BCMA hIgG1 Antibody(Belantamab) (Cat. GM-52396AB) (S1–S4: 1.5-fold from 200 μg/mL; S4–S10: 5-fold from 59.26 μg/mL) for 1 hours, then co-cultured with Membrane Bound H_APRIL(Trimer) HEK-293(Cat. GM-C40503)

for 6 hours. Firefly luciferase activity was then measured using the Luciferase Reporter Assay Kit (Genomeditech) (left Y-axis, relative luminescence units), with inhibition percentages shown on the right Y-axis.

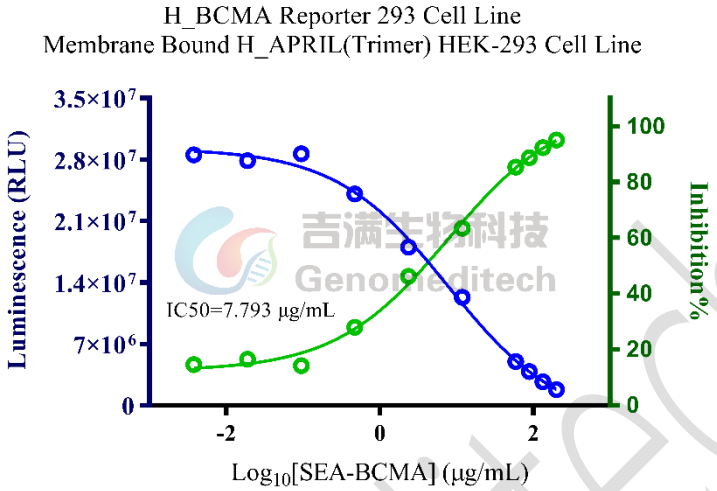
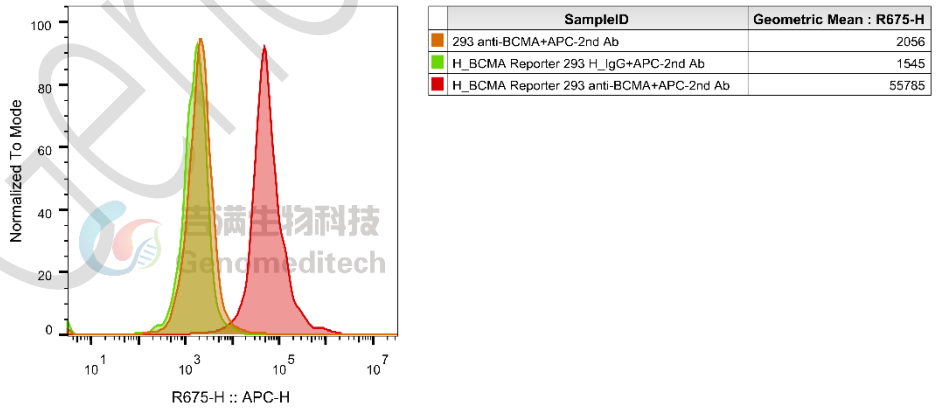


Figure 9 | SEA-BCMA inhibits membrane-bound H_APRIL(Trimer)-induced reporter activity in H_BCMA Reporter 293 cells. The H_BCMA Reporter 293 (Cat. GM-C44705) was seeded at a density of 1.5E4 cells per well in a 96-well plate and incubated overnight. The next day, H_BCMA Reporter 293 Cells were incubated with serial dilutions of Anti-BCMA hIgG1 Antibody(SEA-BCMA) (Cat. GM-49486AB) (S1–S4: 1.5-fold from 200 μg/mL; S4–S10: 5-fold from 59.26 μg/mL) for 1 hours, then co-cultured with Membrane Bound H_APRIL(Trimer) HEK-293(Cat. GM-C40503) for 6 hours. Firefly luciferase activity was then measured using the Luciferase Reporter Assay Kit (Genomeditech) (left Y-axis, relative luminescence units), with inhibition percentages shown on the right Y-axis.



SampleID	Geometric Mean : R675-H
293 anti-BCMA+APC-2nd Ab	2056
H_BCMA Reporter 293 H_IgG+APC-2nd Ab	1545
H_BCMA Reporter 293 anti-BCMA+APC-2nd Ab	55785

Figure 10 | H_BCMA Reporter 293 Cell Line (Cat. GM-C44705) was determined by flow cytometry using Anti-BCMA hIgG1 Antibody(Belantamab) (Cat. GM-52396AB).

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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.

a) 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).
b) 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.
c) 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.
d) Resuspend cell pellet with the recommended recovery medium. And dispense into appropriate culture dishes.
e) Incubate the culture at 37°C in a suitable incubator. A 5% CO_2 in air atmosphere is recommended if using the medium described on this product sheet.

Cell Freezing

Freezing Medium: 90% FBS+10% DMSO

a) Centrifuge at 176 x g for 3 minutes to collect cells.
b) Resuspend the cells in pre-cooled freezing medium and adjust the cell density to 5E6 cells/mL.
c) 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+0.75 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.

a) Subculturing is necessary when the cell density reaches 80%. It is recommended to perform subculturing at a ratio of 1:3 to 1:4 every 2-3 days. Ensure that the density does not exceed 80%, as overcrowding can lead to reduced viability due to compression.
b) Remove and discard culture medium.
c) Briefly rinse the cell layer with PBS to remove all traces of serum that contains trypsin inhibitor.
d) 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).
e) 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.
f) Add 2.0 mL of growth medium to mix well and aspirate cells by gently pipetting.

g) After centrifugation, resuspend the pellet and add appropriate aliquots of the cell suspension to new culture vessels.

h) 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

a) Upon initial thawing, a higher number of dead cells is observed, which is a normal phenomenon. Significant improvement is seen after adaptation. Once the cells reach a stable state, the number of dead cells decreases after subculturing and the cell growth rate becomes stable.

b) Ensure that the cell density does not exceed 80%, as overcrowding may lead to reduced viability due to compression.

Related Products

CD40:CD40L	
H_CD40(TNFRSF5) Reporter 293 Cell Line	H_CD40(TNFRSF5) Reporter Jurkat Cell Line
Cynomolgus_CD40 CHO-K1 Cell Line	Cynomolgus_CD40L CHO-K1 Cell Line
H_CD40(TNFRSF5) CHO-K1 Cell Line	H_CD40(TNFRSF5) HEK-293 Cell Line
H_CD40L CHO-K1 Cell Line	H_CD40L HEK-293 Cell Line
Mouse_CD40L CHO-K1 Cell Line	Rabbit_CD40L NIH-3T3 Cell Line
Anti-CD40 hIgG1 Reference Antibody (Sotibio)	Anti-CD40 hIgG1 Reference Antibody (Tenebio)
Anti-CD40L hIgG1 Reference Antibody (Frebio)	Anti-H_CD40 hIgG1 Antibody(APX005M)
Anti-H_CD40 hIgG1 Antibody(ravagalimab)	Anti-H_CD40L hIgG1 Antibody(dapirolizumab)
Anti-H_CD40L hIgG1 Antibody(frexalimab)	
Biotinylated Human CD40 Protein; His-Avi Tag	Cynomolgus CD40 Protein; His Tag
Human CD40 Protein; His Tag	Human CD40L Protein; His Tag
IFN-α	
IFNα Reporter HEK-293 Cell Line	IFNα Reporter MDCK Cell Line
IFNα 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_BAFFR CHO-K1 Cell Line	H_BAFFR HEK-293 Cell Line
H_BCMA CHO-K1 Cell Line	H_BCMA HEK-293 Cell Line
Membrane Bound H_APRIL(Trimer) HEK-293 Cell Line	
Anti-BAFF hIgG1 Antibody(belimumab)	Anti-BAFFR hIgG1 Antibody(ianalumab)
Anti-BCMA hIgG1 Antibody(Belantamab)	Anti-BCMA hIgG1 Antibody(SEA-BCMA)
Anti-BCMA hIgG4 Antibody(BCMB69)	Anti-CD3E×BCMA hIgG4 Reference Antibody (Tecbio)
Anti-TNFSF13B(BAFF) hIgG1 Reference Antibody (Belibio)	

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 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
CD3	
Jurkat CD3-BsAb Reporter Cell Line	Cynomolgus_CD3 HEK-293 Cell Line
Cynomolgus_CD3E(Membrane Bound ECD) CHO-K1 Cell Line	H_CD3 CHO-K1 Cell Line
H_CD3 HEK-293 Cell Line	H_CD3(TCR V2) CHO-K1 Cell Line
H_CD3(TCR V2) HEK-293 Cell Line	H_CD3(TCRαβ delta V) HEK-293 Cell Line
H_CD3D CD3E KO Jurkat Cell Line	H_CD3E KO Jurkat Cell Line
H_CD3E(Membrane Bound ECD) CHO-K1 Cell Line	Mouse_CD3 HEK-293 Cell Line
Anti-CD19×CD3 hIgG1 Antibody[PIT-565(CD58 K34A)]	Anti-CD3 epsilon hIgG1 Antibody [OKT-3 (muromonab)]
Anti-CD3 hIgG1 Antibody(CH2527)	Anti-CD3×CD20 hIgG1 Bispecific Antibody (Epcobio)
Anti-CD3×FCRL5 hIgG1 Bispecific Antibody(cevostamab)	Anti-CD3E×BCMA hIgG4 Reference Antibody (Tecbio)
Anti-CD3E×DLL3 hIgG1 Bispecific Antibody(Tarlabio)	Anti-CD3E×MUC17 hIgG1 Bispecific Antibody(Vepsitbio)
Anti-mouse CD3ε mIgG2a Antibody(145-2C11)	

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