Genomeditech (Shanghai) Co.,Ltd.
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Product Sheet

H_HVEM PD-L1 aAPC CHO-K1 Cell Line

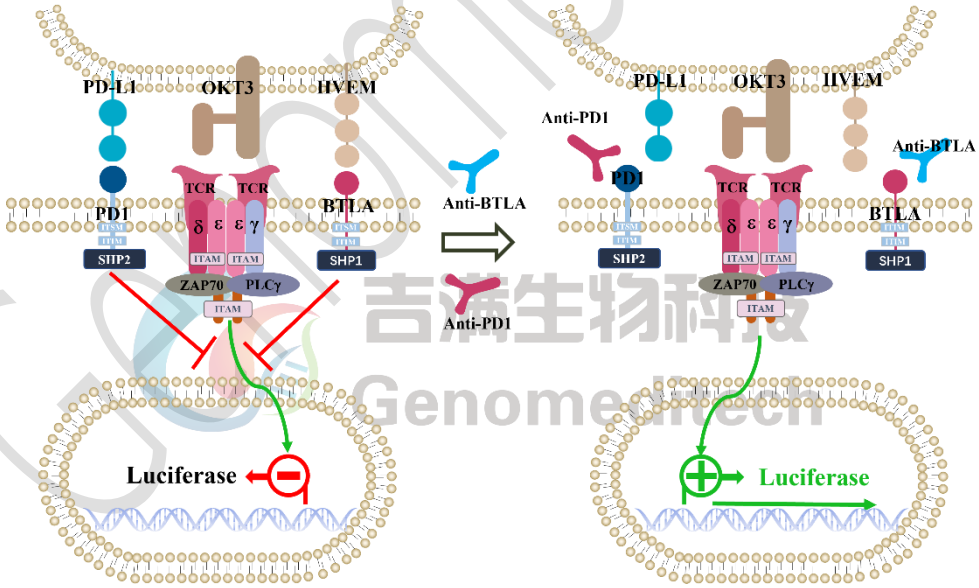
Catalog number: GM-C31561

Version 3.3.1.241120

BTLA (B and T lymphocyte attenuator) and HVEM (herpesvirus entry mediator) are key immune regulatory proteins that form an important regulatory pathway. BTLA, as an inhibitory co-stimulatory receptor, suppresses T cell activation and immune responses when bound to HVEM.

PD-1 (programmed cell death protein 1) is a receptor on activated T cells, B cells, and NK cells that inhibits T cell activation by binding to its ligands PD-L1 and PD-L2. PD-L1, primarily found on tumor and immune cells, promotes immune evasion. The PD-1/PD-L1 pathway is crucial for regulating immune tolerance and inhibiting autoimmune responses, maintaining immune balance.

The H_HVEM PD-L1 aAPC CHO-K1 Cell Line is a clonal stable cell line that constitutively expresses human HVEM and PD-L1 gene. The cell line is co-cultured with the H_BTLA PD-1 Reporter Cell Line. The binding of BTLA to HVEM and PD-1 to PD-L1 both inhibit T cell signaling. By adding Anti-BTLA and Anti-PD1 antibodies, the interactions of BTLA-HVEM and PD-1-PD-L1 are blocked, thereby restoring T cell signaling. The luciferase readout indicates the activation level of the signaling pathway, allowing evaluation of the in vitro effects of BTLA and PD-1 related drugs.



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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	F12K+10% FBS+1% P.S
Growth medium	F12K+10% FBS+1% P.S+4 μg/mL Blasticidin+ 100 μg/mL Hygromycin+4 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.
F12K	BOSTER/PYG0036
Fetal Bovine Serum	Cegrogen biotech/A0500-3010
Pen/Strep	Thermo/15140-122
Blasticidin	Genomeditech/GM-040404
Hygromycin	Genomeditech/GM-040403
Puromycin	Genomeditech/GM-040401
Anti-TNFRSF14(HVEM) hIgG4 Antibody	Genomeditech/GM-49928AB
Anti-H_CD274(PDL1) hIgG1 Antibody(Atezolizumab)	Genomeditech/GM-31740AB

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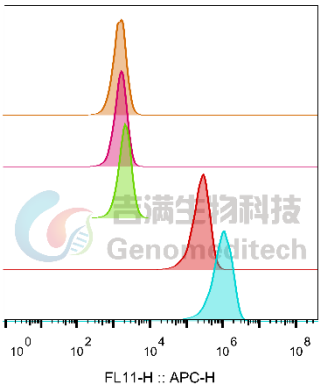
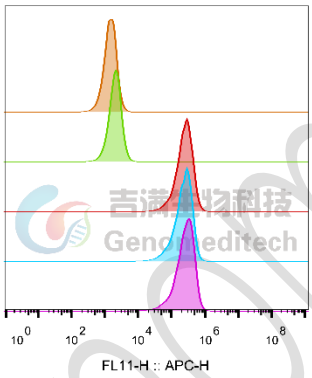
Figures



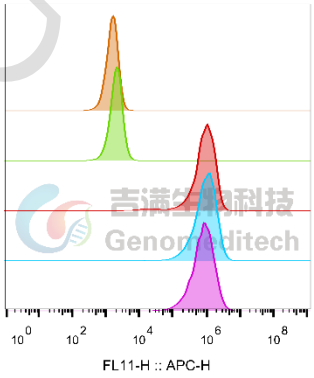
SampleID	Geometric Mean : FL11-H
CHO-K1 anti-HVEM+APC-2nd Ab	1448
CHO-K1 anti-PDL1+APC-2nd Ab	1538
CHO-K1 H_HVEM PD-L1 aAPC H_IgG+APC-2nd Ab	2015
CHO-K1 H_HVEM PD-L1 aAPC anti-HVEM+APC-2nd Ab	230623
CHO-K1 H_HVEM PD-L1 aAPC anti-PDL1+APC-2nd Ab	7.78E5

Figure 1 | H_HVEM PD-L1 aAPC CHO-K1 Cell Line (Cat. GM-C31561) was determined by flow cytometry using Anti-TNFRSF14(HVEM) hIgG4 Antibody (Cat. GM-49928AB), Anti-H_CD274(PDL1) hIgG1 Antibody(Atezolizumab) (Cat. GM-31740AB).



SampleID	Geometric Mean : FL11-H
CHO-K1 anti-HVEM+APC-2nd Ab	1450
CHO-K1 H_HVEM PD-L1 aAPC H_IgG+APC-2nd Ab	2026
CHO-K1 H_HVEM PD-L1 aAPC anti-HVEM+APC-2nd Ab CP5	230806
CHO-K1 H_HVEM PD-L1 aAPC anti-HVEM+APC-2nd Ab CP15	211745
CHO-K1 H_HVEM PD-L1 aAPC anti-HVEM+APC-2nd Ab CP25	240511

Figure 2 | The passage stability of the H_HVEM PD-L1 aAPC CHO-K1 Cell Line (Cat. GM-C31561) was determined by flow cytometry using Anti-TNFRSF14(HVEM) hIgG4 Antibody (Cat. GM-49928AB).



SampleID	Geometric Mean : FL11-H
CHO-K1 anti-PDL1+APC-2nd Ab	1540
CHO-K1 H_HVEM PD-L1 aAPC H_IgG+APC-2nd Ab	2032
CHO-K1 H_HVEM PD-L1 aAPC anti-PDL1+APC-2nd Ab CP5	7.79E5
CHO-K1 H_HVEM PD-L1 aAPC anti-PDL1+APC-2nd Ab CP15	8.87E5
CHO-K1 H_HVEM PD-L1 aAPC anti-PDL1+APC-2nd Ab CP25	7.69E5

Figure 3 | The passage stability of the H_HVEM PD-L1 aAPC CHO-K1 Cell Line (Cat. GM-C31561) was determined by flow cytometry using Anti-H_CD274(PDL1) hIgG1 Antibody(Atezolizumab) (Cat. GM-31740AB).

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Cell Recovery

Recovery Medium: F12K+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: F12K+10% FBS+1% P.S+4 μg/mL Blasticidin+ 100 μg/mL Hygromycin+4 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) Remove and discard culture medium.
b) Briefly rinse the cell layer with PBS to remove all traces of serum that contains trypsin inhibitor.
c) 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 2 to 3 minutes at 37°C).
d) 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.
e) Add 2.0 mL of growth medium to mix well and aspirate cells by gently pipetting.
f) After centrifugation, resuspend the pellet and add appropriate aliquots of the cell suspension to new culture vessels.

g) Incubate cultures at 37°C.

Subcultivation Ratio: A subcultivation ratio of 1:4 - 1:5 is recommended

Medium Renewal: Every 2 to 3 days

Notes

a) After the stabilization of the cell condition, there will be fewer dead cells post-passage, the cell growth rate will tend to stabilize, cell morphology will become uniform, and the cells will appear robust.

Related Products

BTLA:HVEM:LIGHT	
H_BTLA PD-1 Reporter Cell Line	H_BTLA Reporter Cell Line
H_HVEM aAPC CHO-K1 Cell Line	H_HVEM Reporter Jurkat Cell Line
Cynomolgus_BTLA HEK-293 Cell Line	H_BTLA CHO-K1 Cell Line
H_BTLA HEK-293 Cell Line	H_LIGHT(TNFSF14) CHO-K1 Cell Line
H_TNFRSF14(HVEM) CHO-K1 Cell Line	
Anti-BTLA hIgG4 Antibody(22B3)	Anti-BTLA hIgG4 Antibody(Icatolimab)
Anti-TNFRSF14(HVEM) hIgG4 Antibody	Anti-TNFSF14 hIgG4 Antibody

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