

# Product Sheet

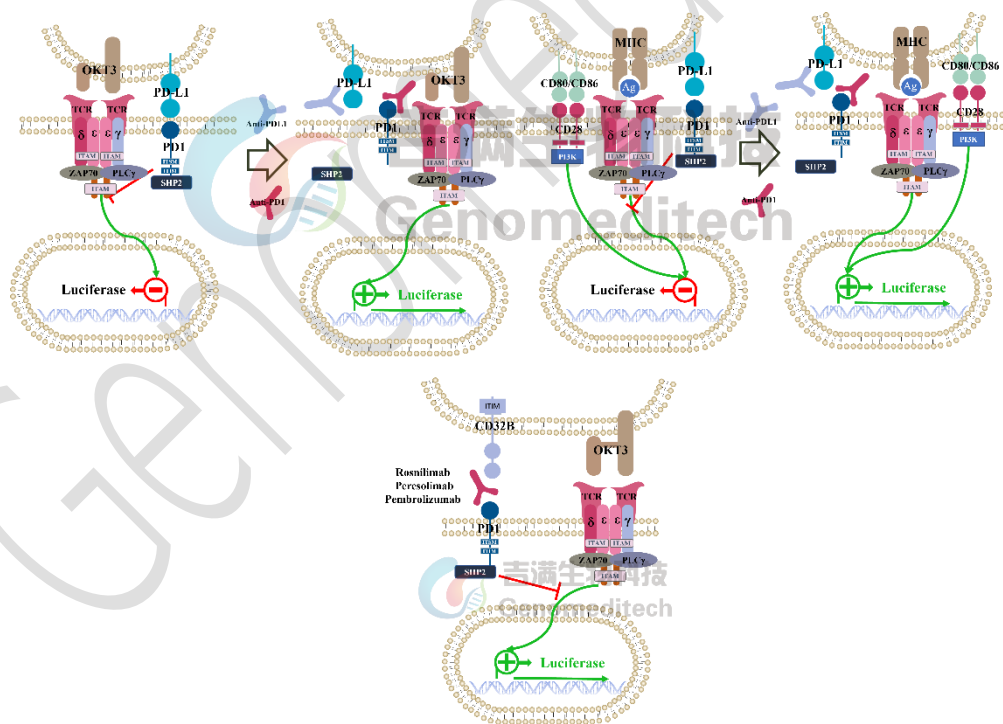
## H\_PD-1 Reporter Jurkat Cell Line

Catalog number: GM-C07928

Version 3.3.1.260126

PD-1 is an immunosuppressive receptor on activated T and B cells, essential for regulating immune responses to tumor antibodies and self-antigens. Its interaction with ligands PD-L1 or PD-L2 inhibits TCR signaling, affecting cell proliferation, transcriptional activation, and cytokine production. Therapeutic antibodies and Fc fusion proteins blocking this interaction have shown promise in cancer clinical trials.

H\_PD-1 Reporter Jurkat Cell Line is a stable clonal Jurkat cell line constructed using lentiviral technology, constitutive expression of the PD-1 gene, along with signal-dependent expression of a luciferase reporter gene. It has three applicable cells: when co-cultured with the aAPC(OKT3) PDL1 CHO-K1 cell line or the H\_PD-L1 Raji cell line, the PD-1/PD-L1 interaction inhibits TCR signaling and luciferase expression. Adding antibodies that block this interaction can relieve the inhibition, restoring TCR signaling and luciferase expression. When co-cultured with the H\_CD32B aAPC CHO-K1 cell line, the addition of PD-1 agonist antibodies allows the antibodies to crosslink with the FcγRIIb receptor, activating PD-1 downstream SHP2-mediated inhibitory signaling, thereby suppressing downstream TCR activation signals.



## 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>	RPMI 1640+10% FBS+1% P.S
<b>Growth medium</b>	RPMI 1640+10% FBS+1% P.S+3.5 µg/mL Blasticidin+0.75 µg/mL Puromycin
<b>Note</b>	None
<b>Freezing Medium</b>	90% FBS+10% DMSO
<b>Growth properties</b>	Suspension
<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

Reagent	Manufacturer/Catalogue No.
RPMI 1640	VivaCell/C3010-0500
Fetal Bovine Serum	ExCell/FSP500
Pen/Strep	Thermo/15140-122
Blasticidin	Genomeditech/ <a href="#">GM-040404</a>
Puromycin	Genomeditech/ <a href="#">GM-040401</a>
aAPC(OKT3) PDL1 CHO-K1 Cell Line	Genomeditech/ <a href="#">GM-C05269</a>
H_PD-L1 Raji Cell Line	Genomeditech/ <a href="#">GM-C03541</a>
H_CD32B aAPC CHO-K1 Cell Line	Genomeditech/ <a href="#">GM-C25754</a>
Staphylococcal Enterotoxin E (SEE)	Genomeditech/GM-H23036
Anti-H_CD274(PDL1) hIgG1 Antibody(Atezolizumab)	Genomeditech/ <a href="#">GM-31740AB</a>
Anti-PD1 hIgG1 Antibody(Rosnilimab)	Genomeditech/ <a href="#">GM-87952AB</a>
Anti-PD1 hIgG4 Antibody(Pembrolizumab)	Genomeditech/ <a href="#">GM-52674AB</a>
Anti-PD1 hIgG1 Antibody(Peresolimab)	Genomeditech/GM-87951AB
Anti-PD1 hIgG4 Reference Antibody (Pembio)	Genomeditech/ <a href="#">GM-87802MAB</a>
GMOne-Step 2.0 Luciferase Reporter Gene Assay Kit	Genomeditech/ <a href="#">GM-040513</a>

## Figures

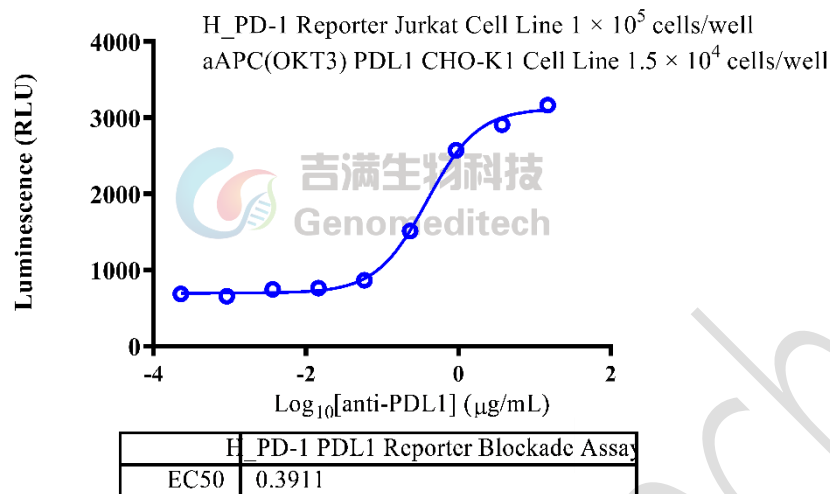


Figure 1 | Response to Anti-H\_CD274(PDL1) hIgG1 Antibody(Atezolizumab). Serial dilutions of the Anti-H\_CD274(PDL1) hIgG1 Antibody (Atezolizumab) (Cat. [GM-31740AB](#)) was incubated with  $1E4$  cells/well of the aAPC(OKT3) PDL1 CHO-K1 Cell Line (Cat. [GM-C05269](#)) in a 96-well plate for 1 hour. Subsequently, H\_PD-1 Reporter Jurkat Cell Line (Cat. GM-C07928) with a concentration of  $1E5$  cells/well was added, and the co-culture proceeded for an additional 16 hours in assay buffer (RPMI 1640 + 1% FBS + 1% P.S). The firefly luciferase activity was measured using the Luciferase Reporter Assay Kit (Genomeditech). The results indicated a maximum fold of approximately [5.1]. Data are shown by drug mass concentration.

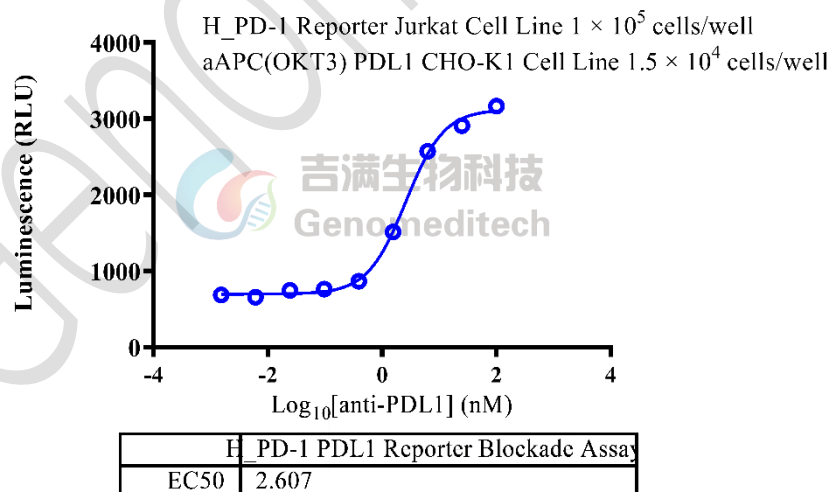


Figure 2 | Response to Anti-H\_CD274(PDL1) hIgG1 Antibody(Atezolizumab). Serial dilutions of the Anti-H\_CD274(PDL1) hIgG1 Antibody (Atezolizumab) (Cat. [GM-31740AB](#)) was incubated with  $1.5E4$  cells/well of the aAPC(OKT3) PDL1 CHO-K1 Cell Line (Cat. [GM-C05269](#)) in a 96-well plate for 1 hour. Subsequently, H\_PD-1 Reporter Jurkat Cell Line (Cat. GM-C07928) with a concentration of  $1E5$  cells/well was added, and the co-culture proceeded for an additional 16 hours in assay buffer (RPMI 1640 + 1% FBS + 1% P.S). The firefly luciferase activity was

measured using the Luciferase Reporter Assay Kit (Genomeditech). The results indicated a maximum fold of approximately [5.1]. Data are shown by drug molar concentration.

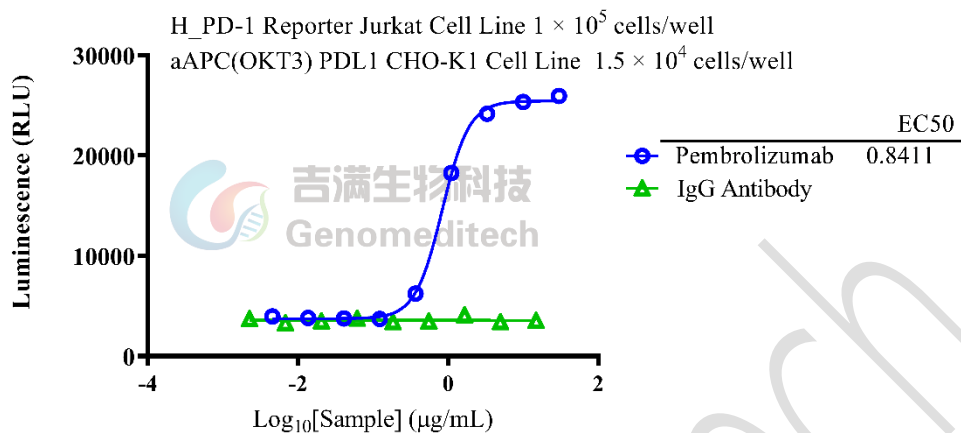


Figure 3 | Response to Anti-PD1 hIgG4 Antibody(Pembrolizumab). aAPC(OKT3) PDL1 CHO-K1 Cell Line (Cat. [GM-C05269](#)) was seeded at a density of 1E4 cells/well in a 96-well plate and incubated overnight. The next day, serial dilutions of the Anti-PD1 hIgG4 Antibody(Pembrolizumab) (Cat. [GM-52674AB](#)), and Human IgG1 Isotype Control were incubated with 1E5 cells/well of the H\_PD-1 Reporter Jurkat Cell Line (Cat. GM-C07928) in a 96-well plate for 1 hour, and then added to the pre-seeded cells. The mixture was incubated for an additional 6 hours. The firefly luciferase activity was measured using the Luciferase Reporter Assay Kit (Genomeditech). The results indicated a maximum fold of approximately [7.3]. Data are shown by drug mass concentration.

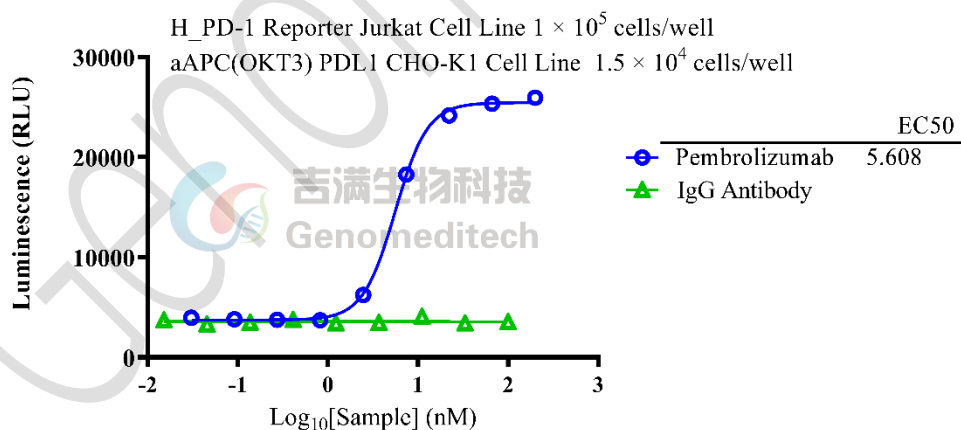


Figure 4 | Response to Anti-PD1 hIgG4 Antibody(Pembrolizumab). aAPC(OKT3) PDL1 CHO-K1 Cell Line (Cat. [GM-C05269](#)) was seeded at a density of 1.5E4 cells/well in a 96-well plate and incubated overnight. The next day, serial dilutions of the Anti-PD1 hIgG4 Antibody(Pembrolizumab) (Cat. [GM-52674AB](#)), and Human IgG1 Isotype Control were incubated with 1E5 cells/well of the H\_PD-1 Reporter Jurkat Cell Line (Cat. GM-C07928) in a 96-well plate for 1 hour, and then added to the pre-seeded cells. The mixture was incubated for an additional 6 hours. The firefly luciferase activity was measured using the Luciferase Reporter Assay Kit (Genomeditech). The results indicated a maximum fold of approximately [7.3]. Data are shown by drug molar concentration.

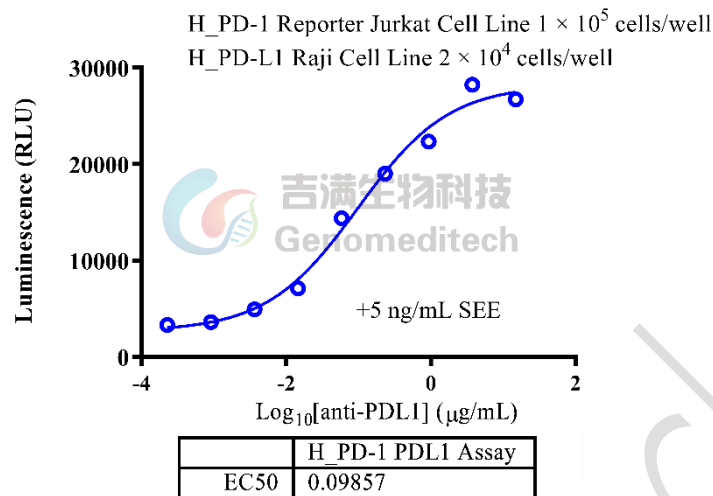


Figure 5 | Response to Anti-H\_CD274(PDL1) hIgG1 Antibody (Atezolizumab). The Atezolizumab (Cat. [GM-31740AB](#)) was serially diluted and incubated with  $2 \times 10^4$  cells/well of the H\_PD-L1 Raji Cell Line in a 96-well plate for 30 minutes. Meanwhile,  $1 \times 10^5$  cells/well of the H\_PD-1 Reporter Jurkat Cell Line (Cat. GM-C07928) was pre-incubated with 0.5 ng/well of SEE in a 96-well plate for 30 minutes. Afterward, the two treated cell mixtures were combined in equal volumes and co-incubated in assay buffer (RPMI 1640 + 1% FBS + 1% P.S.) for 16 hours. The firefly luciferase activity was measured using the Luciferase Reporter Assay Kit (Genomeditech). The results indicated a maximum fold of approximately [8.3]. Data are presented based on drug mass concentration.

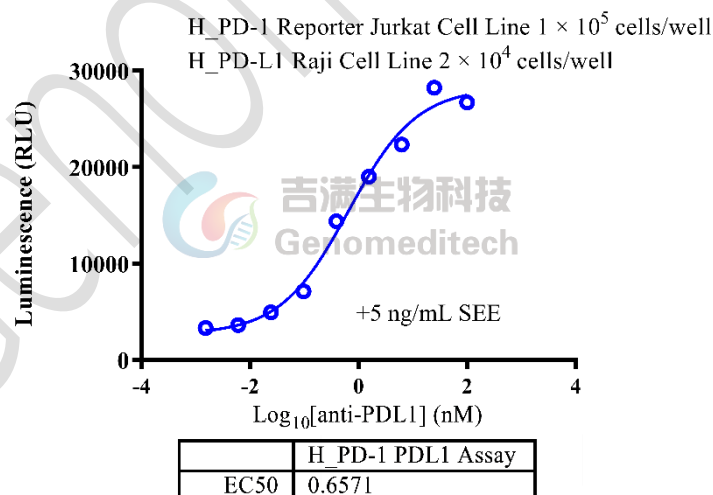


Figure 6 | Response to Anti-H\_CD274(PDL1) hIgG1 Antibody (Atezolizumab). The Atezolizumab (Cat. [GM-31740AB](#)) was serially diluted and incubated with  $2 \times 10^4$  cells/well of the H\_PD-L1 Raji Cell Line in a 96-well plate for 30 minutes. Meanwhile,  $1 \times 10^5$  cells/well of the H\_PD-1 Reporter Jurkat Cell Line (Cat. GM-C07928) was pre-incubated with 0.5 ng/well of SEE in a 96-well plate for 30 minutes. Afterward, the two treated cell mixtures were combined in equal volumes and co-incubated in assay buffer (RPMI 1640 + 1% FBS + 1% P.S.) for 16 hours. The firefly luciferase

activity was measured using the Luciferase Reporter Assay Kit (Genomeditech). The results indicated a maximum fold of approximately [8.3]. Data are presented based on drug molar concentration.

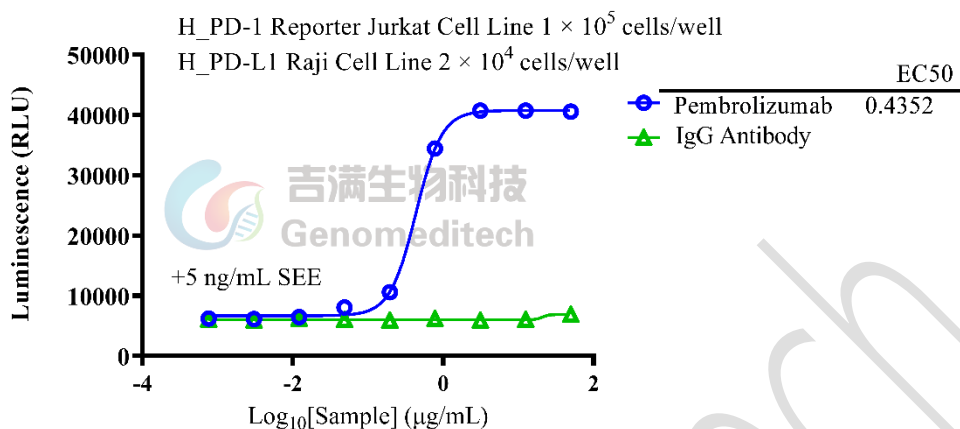


Figure 7 | Response to Anti-PD1 hIgG4 Antibody(Pembrolizumab). The Pembrolizumab (Cat. [GM-52674AB](#)) was serially diluted and incubated with 1E5 cells/well of the H\_PD-1 Reporter Jurkat Cell Line(Cat. GM-C07928) in a 96-well plate for 30 minutes. Meanwhile, 2E4 cells/well of the H\_PD-L1 Raji Cell Line (Cat. [GM-C03541](#)) was pre-incubated with 0.5 ng/well of SEE in a 96-well plate for 30 minutes. Afterward, the two treated cell mixtures were combined in equal volumes and co-incubated in assay buffer for 16 hours. IgG Isotype is the control. The firefly luciferase activity was measured using the Luciferase Reporter Assay Kit (Genomeditech). The results indicated a maximum fold of approximately [6.6]. Data are presented based on drug mass concentration.

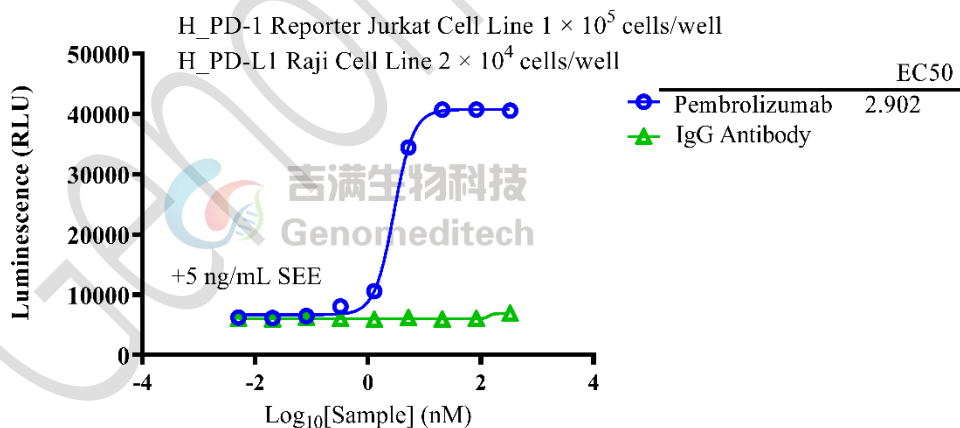


Figure 8 | Response to Anti-PD1 hIgG4 Antibody(Pembrolizumab). The Pembrolizumab (Cat. [GM-52674AB](#)) was serially diluted and incubated with 1E5 cells/well of the H\_PD-1 Reporter Jurkat Cell Line(Cat. GM-C07928) in a 96-well plate for 30 minutes. Meanwhile, 2E4 cells/well of the H\_PD-L1 Raji Cell Line (Cat. [GM-C03541](#)) was pre-incubated with 0.5 ng/well of SEE in a 96-well plate for 30 minutes. Afterward, the two treated cell mixtures were combined in equal volumes and co-incubated in assay buffer for 16 hours. IgG Isotype is the control. The firefly luciferase activity was measured using the Luciferase Reporter Assay Kit (Genomeditech). The results indicated a maximum fold of approximately [6.6]. Data are presented based on drug molar concentration.



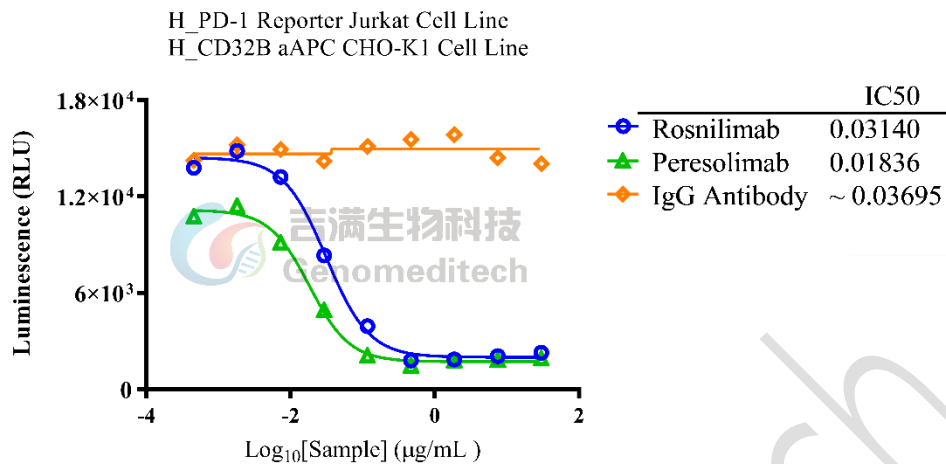


Figure 9 | Response to Rosnilimab and Peresolimab. Serial dilutions of the Rosnilimab and Peresolimab was incubated with 1E4 cells/well of the H\_CD32B aAPC CHO-K1 Cell Line (Cat. [GM-C25754](#)) in a 96-well plate for 1 hour. Subsequently, H\_PD-1 Reporter Jurkat Cell Line (Cat. GM-C07928) with a concentration of 1E5 cells/well was added, and the co-culture proceeded for an additional 6 hours in assay buffer (RPMI 1640 + 1% FBS + 1% P.S). The firefly luciferase activity was measured using the Luciferase Reporter Assay Kit (Genomeditech). The results indicated maximum blocking folds of approximately [5.8] and [5.7], respectively. Data are shown by drug mass concentration.

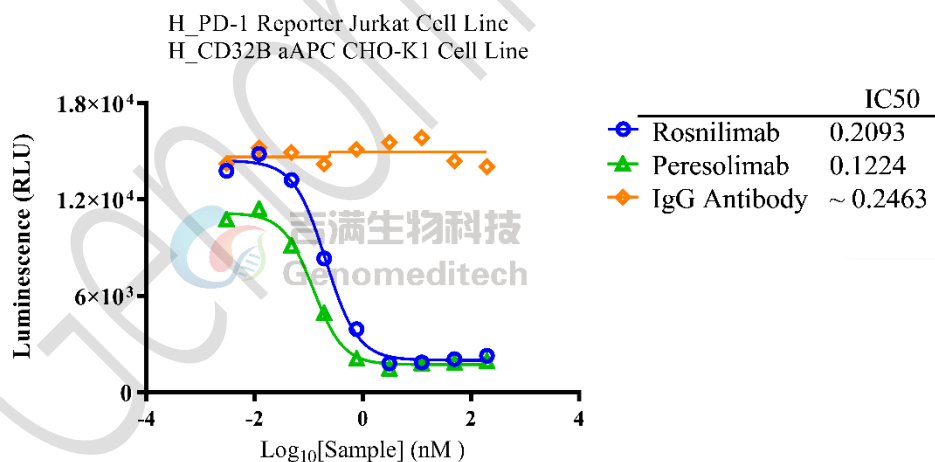


Figure 10 | Response to Rosnilimab and Peresolimab. Serial dilutions of the Rosnilimab and Peresolimab was incubated with 1E4 cells/well of the H\_CD32B aAPC CHO-K1 Cell Line (Cat. [GM-C25754](#)) in a 96-well plate for 1 hour. Subsequently, H\_PD-1 Reporter Jurkat Cell Line (Cat. GM-C07928) with a concentration of 1E5 cells/well was added, and the co-culture proceeded for an additional 6 hours in assay buffer (RPMI 1640 + 1% FBS + 1% P.S). The firefly luciferase activity was measured using the Luciferase Reporter Assay Kit (Genomeditech). The results indicated maximum blocking folds of approximately [5.8] and [5.7]. Respectively. Data are shown by drug molar concentration.

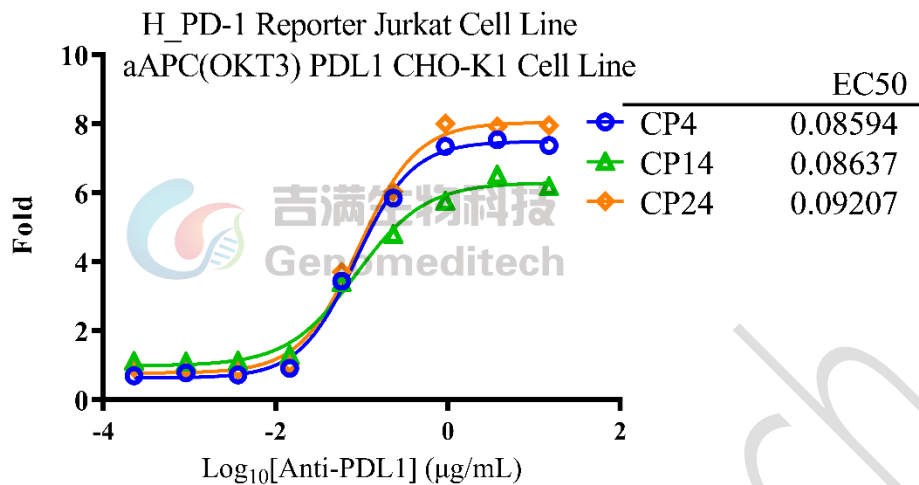


Figure 11 | The passage stability of response to Anti-H\_CD274(PDL1) hIgG1 Antibody(Atezolizumab). Serial dilutions of the Atezolizumab(Cat. [GM-31740AB](#)) were incubated with 1.5E4 cells/well of the aAPC(OKT3) PDL1 CHO-K1 Cell Line (Cat. [GM-C05269](#)) in a 96-well plate for 1 hour in assay buffer (RPMI 1640 + 1% FBS + 1% P.S). Subsequently, the passage 4, 14, and 24 of H\_PD-1 Reporter Jurkat Cell Line (Cat. [GM-C07928](#)) at a concentration of 1E5 cells/well was added, and the coculture proceeded for an additional 16 hours. The firefly luciferase activity was measured using the Luciferase Reporter Assay Kit (Genomeditech). Data are shown by drug mass concentration.

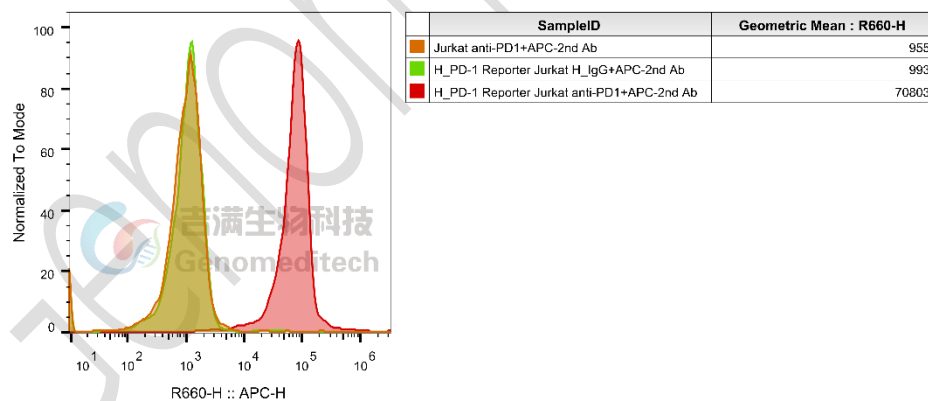


Figure 12 | H\_PD-1 Reporter Jurkat Cell Line (Cat. [GM-C07928](#)) was determined by flow cytometry using Anti-PD1 hIgG1 Antibody (In house).

## Cell Recovery

Recovery Medium: RPMI 1640+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  $-70^{\circ}\text{C}$ . Storage at  $-70^{\circ}\text{C}$  will result in loss of viability.

- Thaw the vial by gentle agitation in a  $37^{\circ}\text{C}$  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  $176 \times g$  for 5 minutes. Discard supernatant.
- Resuspend cell pellet with the recommended complete medium. And dispense the suspension into 1 - 2 T-25 culture flasks.
- Incubate the culture at  $37^{\circ}\text{C}$  in a suitable incubator. A 5%  $\text{CO}_2$  in air atmosphere is recommended if using the medium described on this product sheet.

## Cell Freezing

Freezing Medium: 90% FBS+10% DMSO

- Centrifuge at  $176 \times g$  for 3 minutes to collect cells.
- Resuspend the cells in pre-cooled freezing medium and adjust the cell density to  $5 \times 10^6$  cells/mL.
- Aliquot 1 mL into each vial.
- Place the vial in a controlled-rate freezing container and store at  $-80^{\circ}\text{C}$  for at least 1 day, then transfer to liquid nitrogen as soon as possible.

## Cell passage

Growth medium: RPMI 1640+10% FBS+1% P.S+3.5  $\mu\text{g/mL}$  Blasticidin+0.75  $\mu\text{g/mL}$  Puromycin

Approximately 48-72 hours after the initial thawing, the cells can be passaged for the first time. After this initial passage, the culture medium can be adjusted to growth medium supplemented with antibiotics. If cells are not passaged within 48 hours, it is recommended to add some fresh recovery medium and place the flask horizontally.

- When the cell density reaches  $1.5 - 2 \times 10^6$  cells/mL, subculture the cells. Do not allow the cell density to exceed  $2 \times 10^6$  cells/mL.
- It is recommended to use T-25 flasks for subculturing.
- These cells are suspension cells, and it is recommended to use the "half-medium change" method to maintain optimal cell conditions during passaging.
- During passaging, you can directly add fresh growth medium to the culture flask, gently pipette to resuspend the cells, and then transfer the cell suspension to a new T-25 flask for continued culture.

**Subcultivation Ratio: Maintain cultures at a cell concentration between  $3 \times 10^5$  and  $1 \times 10^6$  viable cells/mL.**

**Medium Renewal: Every 2 to 3 days**

## Notes

- These cells are sensitive to density, so please ensure that the cell density is maintained within an appropriate range during culture and subculturing.
- During the first passage, pay attention to the nutrient supply; if not subculturing, make sure to add fresh recovery medium every other day as needed.

## Related Products

PD-1:PD-L1(B7-H1):PDL2	
Mouse_PDL1 KO LLC1 Cell Line	Mouse_PDL1 KO MC38 Cell Line
aAPC(OKT3) PDL1 CHO-K1 Cell Line	H_PD1 SHP1 Reporter Jurkat Cell line
H_PDCD1LG2(PDL2) aAPC CHO-K1 Cell Line	Mouse PDL1 aAPC CHO-K1 Cell Line
Mouse_PD-1 Reporter Jurkat Cell Line	Canine_PD-1 CHO-K1 Cell Line
Canine_PD-1 HEK-293 Cell Line	Cynomolgus_PD1 CHO-K1 Cell Line
Cynomolgus_PD-L1 HEK-293 Cell Line	H_CD274(PD-L1) CHO-K1 Cell Line
H_CD274(PD-L1) MC38 Cell Line	H_PDCD1(PD-1) CHO-K1 Cell Line
H_PDCD1(PD-1) CHO-K1 Cell Line (Low Expression)	H_PDCD1(PD-1) HEK-293 Cell Line
H_PDCD1LG2(PDL2) CHO-K1 Cell Line	H_PD-L1 HEK-293 Cell Line
H_PDL1 LLC1(mouse_PDL1 KO) Cell Line	H_PDL1 LLC1(mouse_PDL1 KO) Cell Line
H_PDL1 MC38(mouse_PDL1 KO) Cell Line	H_PD-L1 Raji Cell Line
M_PDCD1(PD-1) CHO-K1 Cell Line	
Anti-Canine_PD1 mIgG2a Antibody(4F12-E6)	Anti-CTLA-4/PD-1 hIgG1 Bispecific Antibody(Cadonilimab)
Anti-CTLA4×PD-1 hIgG1 Reference Antibody (Cadbio)	Anti-H_CD274(PDL1) hIgG1 Antibody(Atezolizumab)
Anti-H_PDCD1(PD1) hIgG1 Antibody(Budigalimab)	Anti-H_PDCD1LG2 mIgG1 Antibody(3G2)
Anti-H_PDL1 hIgG1 Reference Antibody(Atezbio)	Anti-mouse PD1 RIgG2a Antibody(RMP1-14)
Anti-mouse PD-L1 mIgG1 Antibody(10F.9G2)	Anti-Mouse_PD1 mIgG1 Antibody(29F.1A12)
Anti-mouse_PD1 mIgG1 Antibody(RMP1-14)	Anti-Mouse_PD1×VEGF hIgG1 Bispecific Antibody
Anti-PD1 hIgG1 Reference Antibody (Perbio)	Anti-PD1 hIgG1 Reference Antibody(Rosnbio)
Anti-PD1 hIgG4 Antibody(Pembrolizumab)	Anti-PD1 hIgG4 Reference Antibody (Nivbio)
Anti-PD1 hIgG4 Reference Antibody (Pembio)	Anti-PD1 hIgG4 Reference Antibody (Sintbio)
Anti-PD-1 hIgG4 Reference Antibody (Torbio)	Anti-PD1 hIgG4 Reference Antibody(Cambio)
Anti-PD-1 hIgG4 Reference Antibody(Tisbio)	Anti-PD1-IL2v Fusion hIgG1 Antibody(2149)
Anti-PD1-IL2v Fusion hIgG1 Antibody(KY-0118)	Anti-PD-L1 hIgG1 Reference Antibody(Avebio)
Anti-PDL1 hIgG4 Reference Antibody(Adebio)	Anti-PD-L2 hIgG1 Antibody(Hz25G4-1.1)
Anti-VEGF×PD1 hIgG1 Reference Antibody (Ivobio)	Anti-VEGF×PD-L1 hIgG1 Bispecific Antibody (Pumibio)
Anti-VEGF×PD-L1 hIgG1 Bispecific Antibody (pumitamig)	
Biotinylated Human PD1 Protein; His-Avi Tag	Biotinylated Human PDL1 Protein; His-Avi Tag
Canine PD1 Protein; hFc Tag	Cynomolgus PDL1 Protein; His Tag
Human PD1 Protein; hFc Tag	Human PD1 Protein; His Tag

Human PDL1 Protein; His Tag	Human PDL1 Protein; mFc Tag
Human PDL2 Protein; mFc Tag	Mouse PDL1 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.
- Users and their contractors engaged for their benefit may use this material and its derivatives only within the agreed research scope; modification of the material is not permitted, nor may it be distributed, sold, transferred, or otherwise provided to any other entity (including affiliates).
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