

# Product Sheet

## H\_HLA-G1 OKT3 CHO-K1 Cell Line

Catalog number: GM-C16834

Version 3.3.1.241206

HLA-G1 is a non-classical MHC class I molecule with immunosuppressive functions. It binds to inhibitory receptors such as LILRB1 (ILT2) and LILRB2 (ILT4) to suppress the activity of effector immune cells, including T cells, NK cells, and B cells, thereby inducing immune tolerance. HLA-G1 plays a critical role in immune checkpoint regulation, promoting T cell anergy and the differentiation of regulatory T cells (Tregs), ultimately suppressing immune responses.

OKT3 is an anti-CD3 monoclonal antibody that specifically binds to the CD3 molecule on the surface of T cells. CD3 is a component of the T cell receptor (TCR) complex and is involved in T cell activation and signal transduction. By binding to CD3, OKT3 strongly activates T cells, inducing their proliferation and cytokine release. However, in certain cases, prolonged stimulation by OKT3 can lead to T cell exhaustion or apoptosis.

H\_HLA-G1 OKT3 CHO-K1 Cell Line is a clonal stable CHO-K1 cell line that constitutively expresses the human HLA-G1 gene, human B2M gene and OKT3 gene, constructed using lentiviral technology.

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## 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>	F12K+10% FBS+1% P.S
<b>Growth medium</b>	F12K+10% FBS+1% P.S+4 $\mu\text{g/mL}$ Blasticidin+100 $\mu\text{g/mL}$ Hygromycin+4 $\mu\text{g/mL}$ 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>
Fetal Bovine Serum	Cegrogen biotech/A0500-3010
Pen/Strep	Thermo/15140-122
Blasticidin	Genomeditech/ <a href="#">GM-040404</a>
Hygromycin	Genomeditech/ <a href="#">GM-040403</a>
Puromycin	Genomeditech/ <a href="#">GM-040401</a>
Anti-H_HLA-G1 hIgG1 Antibody(38373)	Genomeditech/ <a href="#">GM-28208AB</a>

## Figures

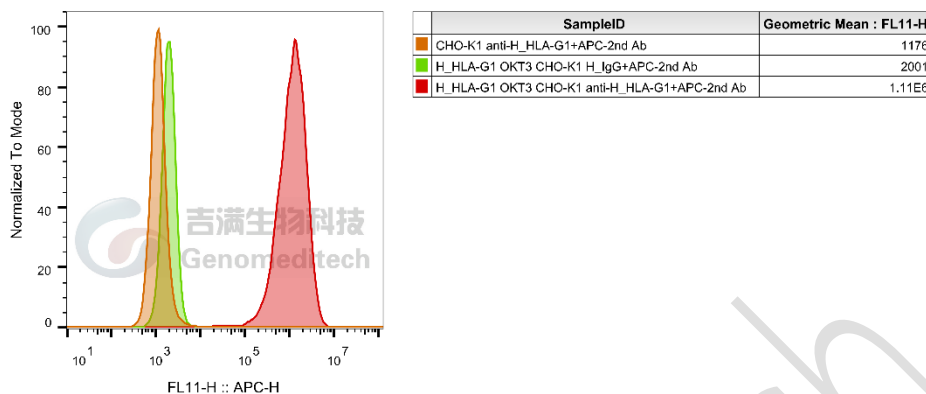


Figure 1 | H\_HLA-G1 OKT3 CHO-K1 Cell Line (Cat. GM-C16834) was determined by flow cytometry using Anti-H\_HLA-G1 hIgG1 Antibody(38373) (Cat. GM-28208AB).

## 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.

- 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: 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.

- 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 2 to 3 minutes 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 1:4 - 1:5 is recommended**

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

## Notes

- 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

HLA-G	
<a href="#">H_HLA-G1 CHO-K1 Cell Line</a>	<a href="#">H_HLA-G1 HEK-293 Cell Line</a>
<a href="#">H_HLA-G1 MC38 Cell Line</a>	<a href="#">H_HLA-G1 PDL1 MC38(mouse_PDL1 KO) Cell Line</a>
<a href="#">H_HLA-G1 SK-OV3 Cell Line</a>	<a href="#">Rhesus_MAMU-AG HEK-293 Cell Line</a>
<a href="#">Anti-H_HLA-G1 hIgG1 Antibody(38373)</a>	

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