

# **Product Sheet**

# H\_NKp30 Reporter Jurkat Cell Line

Catalog number: GM-C29776

Version 3.3.1.241121

Natural killer (NK) cells are an important component of the innate immune system. Among the four known natural cytotoxicity receptors, three are constitutively expressed in NK cells (NKp30, NKp46, and NKp80), while NKp44 is only found on the surface of activated NK cells. NKp30 is expressed on nearly all human NK cells. This NCR has been shown to play a crucial role in the crosstalk between NK cells and dendritic cells (DC) by promoting the maturation of immature DCs and enhancing cytotoxicity. When NKp30 binds to an activating antibody or ligand, it transmits a strong activation signal to the cell, and the signaling of the NCR occurs through the ITAM motifs of FccRIy and CD3ζ in NKp30.

H\_NKp30 Reporter Jurkat Cell Line is a clonal stable Jurkat cell line constitutively expressing the Nkp30 and FCER1G, along with signal-dependent expression of a luciferase reporter gene. The addition of coated NKp30 antibody agonists stimulates NKp30 to bind with CD3 $\zeta$  and Fc $\epsilon$ RI $\gamma$ , activating downstream reporter genes and inducing luciferase expression. This system can be used to evaluate the in vitro effects of antibodies targeting NKp30.





# 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	RPMI 1640+10% FBS+1% P.S		
Growth medium	RPMI 1640+10% FBS+1% P.S+3.5 µg/mL Blasticidin+400 µg/mL G418+0.75 µg/mL Puromycin		
Note	None		
Freezing Medium	90% FBS+10% DMSO		
Growth properties	Suspension		
Growth Conditions	37°C, 5% CO <sub>2</sub>		
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			

# **Materials**

Reagent	Manufacturer/Catalogue No.
RPMI 1640	VivaCell/C3010-0500
Fetal Bovine Serum	Cegrogen biotech/A0500-3010
Pen/Strep	Thermo/15140-122
Blasticidin	Genomeditech/GM-040404
G418	Genomeditech/GM-040402
Puromycin	Genomeditech/GM-040401
Clear Flat-Bottom Immuno Nonsterile 96-Well Plates	Thermo/442404
Anti-NKP30 hIgG1 Antibody(BGA-1833)	Genomeditech/GM-49311AB
Anti-NKP30 hIgG1 Antibody(BJM-0411)	Genomeditech/GM-59269AB
Human IgG1 Isotype Control(Anti-RSV)	Genomeditech/GM-47471AB
GMOne-Step Luciferase Reporter Gene Assay Kit	Genomeditech/GM-040503

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### Figures



Figure 1 | Response to Anti-NKP30 hIgG1 Antibody. H\_NKp30 Reporter Jurkat Cell Line (Cat. GM-C29776) at a concentration of 1E5 cells/well in a 96-well format. The wells were coated overnight with serial dilutions of Anti-NKP30 hIgG1 Antibody (BGA-1833) (Cat. GM-49311AB), Anti-NKP30 hIgG1 Antibody (BJM-0411) (Cat. GM-59269AB) and Human IgG1 Isotype Control (Anti-RSV) (Cat. GM-47471AB) in assay buffer (RPMI 1640+1% FBS+1% P.S). After coating, the cells were added and incubated for 16 hours. The firefly luciferase activity was measured using the GMOne-Step Luciferase Reporter Gene Assay Kit (Cat. GM-040503). The maximum induction fold of BGA-1833 and BJM-0411 was approximately [91.8] and [127.1]. Data are shown by drug mass concentration.



Figure 2 | Response to Anti-NKP30 hIgG1 Antibody. H\_NKp30 Reporter Jurkat Cell Line (Cat. GM-C29776) at a concentration of 1E5 cells/well in a 96-well format. The wells were coated overnight with serial dilutions of Anti-NKP30 hIgG1 Antibody (BGA-1833) (Cat. GM-49311AB), Anti-NKP30 hIgG1 Antibody (BJM-0411) (Cat. GM-59269AB) and Human IgG1 Isotype Control (Anti-RSV) (Cat. GM-47471AB) in assay buffer (RPMI 1640+1% FBS+1% P.S). After coating, the cells were added and incubated for 16 hours. The firefly luciferase activity was measured using the GMOne-Step Luciferase Reporter Gene Assay Kit (Cat. GM-040503). The maximum induction fold of BGA-1833 and BJM-0411 was approximately [91.8] and [127.1]. Data are shown by drug molar concentration.

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# H\_NKp30 Reporter Jurkat Cell Line 1×10<sup>5</sup> cells/well P5/P15/P25

Figure 3 | The passage stability of response to Anti-NKP30 hIgG1 Antibody(BGA-1833). The passage 5, 15 and 25 of H NKp30 Reporter Jurkat Cell Line (Cat. GM-C29776) at a concentration of 1E5 cells/well in a 96-well format. The wells were coated overnight with serial dilutions of Anti-NKP30 hIgG1 Antibody (BGA-1833) (Cat. GM-49311AB) in assay buffer (RPMI 1640 + 1% FBS + 1% P.S). After coating, the cells were added and incubated for 16 hours. The firefly luciferase activity was measured using the GMOne-Step Luciferase Reporter Gene Assay Kit (Cat. GM-040503). Data are shown by drug mass concentration.



Figure 4 | The passage stability of response to Anti-NKP30 hIgG1 Antibody(BGA-1833). The passage 5, 15 and 25 of H\_NKp30 Reporter Jurkat Cell Line (Cat. GM-C29776) at a concentration of 1E5 cells/well in a 96-well format. The wells were coated overnight with serial dilutions of Anti-NKP30 hIgG1 Antibody (BGA-1833) (Cat. GM-49311AB) in assay buffer (RPMI 1640 + 1% FBS + 1% P.S). After coating, the cells were added and incubated for 16 hours. The firefly luciferase activity was measured using the GMOne-Step Luciferase Reporter Gene Assay Kit (Cat. GM-040503). Data are shown by drug molar concentration.

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SampleID	Geometric Mean : FL11-H		
Jukat anti-NKp30+APC-2nd Ab	1417		
H_NKp30 reporter Jukat H_IgG+APC-2nd Ab	1822		
H_NKp30 reporter Jukat anti-NKp30+APC-2nd Ab	25745		

Figure 5 | H\_NKp30 Reporter Jurkat Cell Line (Cat. GM-C29776) was determined by flow cytometry using Anti-NKP30 hJgG1 Antibody(BGA-1833) (Cat. GM-49311AB).

#### **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  $-70^{\circ}$ C. Storage at  $-70^{\circ}$ 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 complete medium. And dispense the suspension into 1 2 T-25 culture flasks.
- e) 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

- 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.

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### Cell passage

Growth medium: RPMI 1640+10% FBS+1% P.S+3.5 µg/mL Blasticidin+400 µg/mL G418+0.75 µg/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.

- a) When the cell density reaches 1.5 2E6 cells/mL, subculture the cells. Do not allow the cell density to exceed 2E6 cells/mL.
- b) It is recommended to use T-25 flasks for subculturing.
- c) These cells are suspension cells, and it is recommended to use the "half-medium change" method to maintain optimal cell conditions during passaging.
- d) 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 concentraion between 3E5 and 1E6 viable cells/mL.

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

#### Notes

- a) These cells are sensitive to density, so please ensure that the cell density is maintained within an appropriate range during culture and subculturing.
- b) 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**

NKp46			
H_Nkp46 Reporter Jurkat Cell Line	Cynomolgus_Ncr1(NKp46) HEK-293 Cell Line		
H_Ncr1(NKp46) CHO-K1 Cell Line	H_Ncr1(NKp46) HEK-293 Cell Line		
Mouse_Ncr1(NKp46) CHO-K1 Cell Line	Rhesus_Ncr1(NKp46) CHO-K1 Cell Line		
Rhesus_Ncr1(NKp46) HEK-293 Cell Line			
Anti-NCR1(NKP46) hIgG1 Antibody(A26-BhlgG1)			
NKP30:B7-H6			
Cynomolgus_NCR3LG1(B7-H6) CHO-K1 Cell Line	Cynomolgus_NKP30 CHO-K1 Cell Line		
H_NCR3LG1(B7-H6) CHO-K1 Cell Line	H_NCR3LG1(B7-H6) HEK-293 Cell Line		
H_NKP30 CHO-K1 Cell Line	H_NKP30 HEK-293 Cell Line		
Anti-NKP30 hIgG1 Antibody(BGA-1833)	Anti-B7-H6 hIgG1 Antibody(BI-765049)		
Biotinylated Cynomolgus NCR3LG1(B7-H6) Protein; His-Avi Tag	Biotinylated Human NCR3LG1(B7-H6) Protein; His-Avi Tag		
Cynomolgus NCR3LG1(B7-H6) Protein; His Tag	Human NCR3(NKP30) Protein; His Tag		
Human NCR3LG1(B7-H6) Protein; His Tag			

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