Genomeditech (Shanghai) Co.,Ltd.
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Product Sheet

H_Vγ9Vδ2(G115) Reporter Jurkat(TCRαβ KO) Cell Line

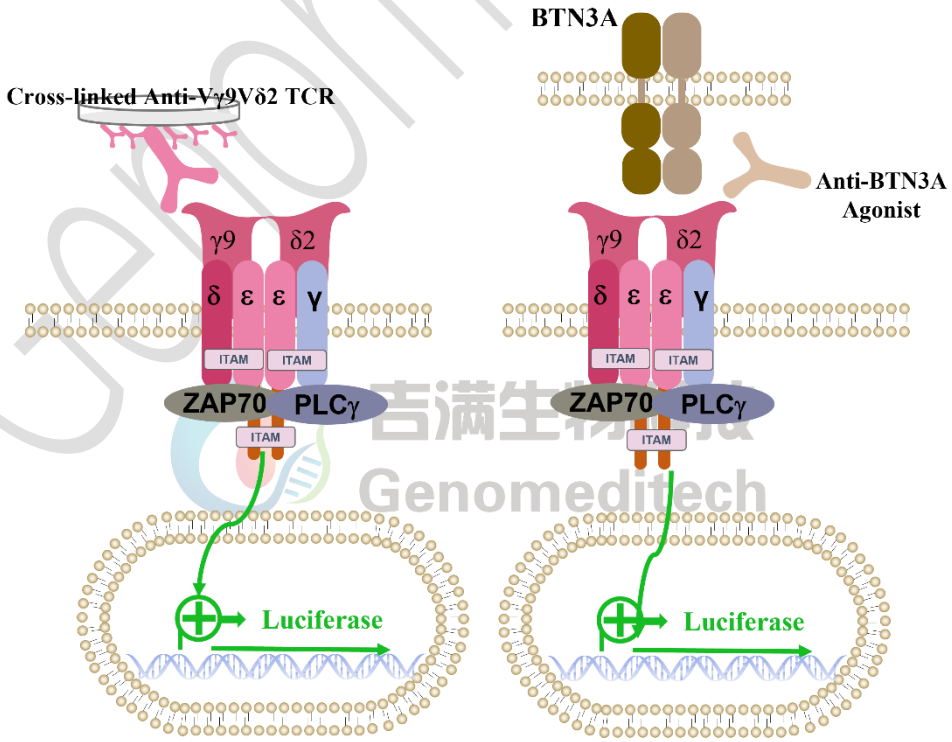
Catalog number: GM-C28019

Version 3.3.1.250116

α β T cells recognize "non-self" or tumor neoantigen peptides via their TCR and constitute 65%-70% of T cells. In contrast, γδ T cells, making up 0.5%-5% of T lymphocytes, have TCRs composed of γ and δ chains and are mainly found in epithelial and mucosal tissues. They can recognize antigens without MHC restriction and effectively kill tumors and pathogens by activating through non-peptide phosphoantigens (PAgs). Monoclonal antibodies against BTN3A1 have been developed to specifically activate Vγ9Vδ2 T cells independently of PAgs.

Binding of BTN3A antibodies to Jurkat cells or Vγ9Vδ2 TCR antibodies to Vγ9Vδ2 activates downstream signaling pathways, including ZAP-70 and LAT, which activate transcription factors like MAPK and NF-κB, promoting cell proliferation and cytokine production. Vγ9Vδ2 T cells can further enhance their function through cytokines (IL-2, IL-15) and costimulatory molecules (CD28, 4-1BB).

H_Vγ9Vδ2(G115) Reporter Jurkat(TCRαβ KO) Cell Line is a clonal stable Jurkat cell line constructed using lentiviral technology, constitutive expression of the Vγ9Vδ2 gene and knockout of TCRαβ, along with signal-dependent expression of a luciferase reporter gene. 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 Vγ9Vδ2(G115).



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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	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+200 μg/mL Hygromycin+0.75 μg/mL Puromycin
Note	None
Freezing Medium	90% FBS+10% DMSO
Growth properties	Suspension
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.
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
Hygromycin	Genomeditech/GM-040403
Puromycin	Genomeditech/GM-040401
Clear Flat-Bottom Immuno Nonsterile 96-Well Plates	Thermo/442404
Anti-Vγ9Vδ2 TCR hIgG1 Antibody	Genomeditech/GM-75325AB
Anti-BTN3A1 hIgG1 Antibody(mAb1)	Genomeditech/GM-52838AB
Anti-CD3 epsilon hIgG1 Antibody [OKT-3 (muromonab)]	Genomeditech/GM-51478AB
GMOne-Step Luciferase Reporter Gene Assay Kit	Genomeditech/GM-040503

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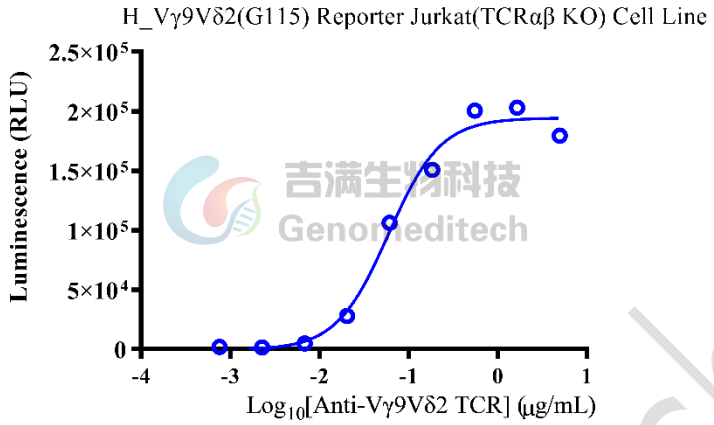
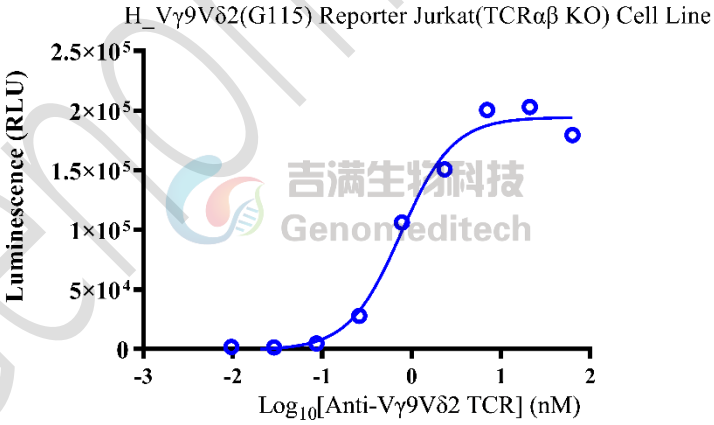
Figures



	H_Vγ9Vδ2(G115)(TCR KO) Reporter
EC50	0.06006

Figure 1 | Response to Anti-Vγ9Vδ2 TCR hIgG1 Antibody. The H_Vγ9Vδ2(G115) Reporter Jurkat(TCRαβ KO) Cell Line (Cat. GM-C28019) at a concentration of 1E5 cells/well in a 96-well format. The wells were coated overnight with serial dilutions of Anti-Vγ9Vδ2 TCR hIgG1 Antibody (Cat. GM-75325AB) in assay buffer (RPMI 1640 + 1% FBS + 1% P.S). After coating, the cells were added and incubated for 24 hours. The firefly luciferase activity was measured using the GMOne-Step Luciferase Reporter Gene Assay Kit (Cat. GM-040503). The maximum induction fold was approximately [104.9]. Data are shown by drug mass concentration.



	H_Vγ9Vδ2(G115)(TCR KO) Reporter
EC50	0.7602

Figure 2 | Response to Anti-Vγ9Vδ2 TCR hIgG1 Antibody. The H_Vγ9Vδ2(G115) Reporter Jurkat(TCRαβ KO) Cell Line (Cat. GM-C28019) at a concentration of 1E5 cells/well in a 96-well format. The wells were coated overnight with serial dilutions of Anti-Vγ9Vδ2 TCR hIgG1 Antibody (Cat. GM-75325AB) in assay buffer (RPMI 1640 + 1% FBS + 1% P.S). After coating, the cells were added and incubated for 24 hours. The firefly luciferase activity was measured using the GMOne-Step Luciferase Reporter Gene Assay Kit (Cat. GM-040503). The maximum induction fold was approximately [104.9]. Data are shown by drug molar concentration.

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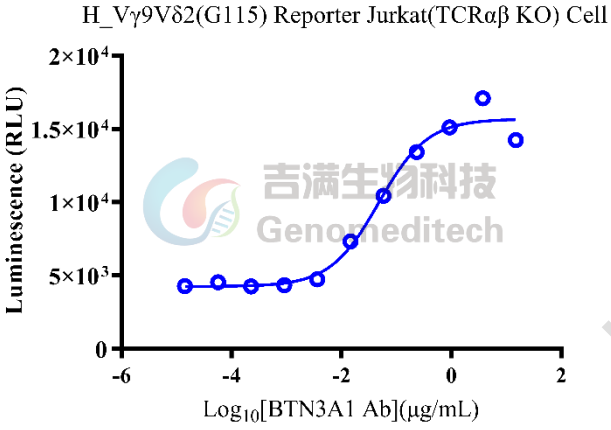
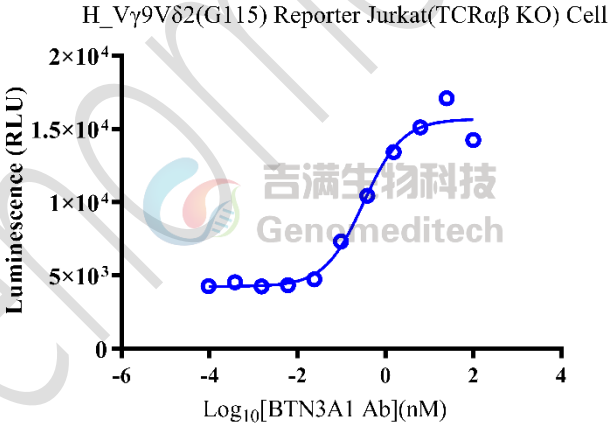


	H_Vγ9Vδ2(G115)(TCR KO) Reporter
EC50	0.04805

Figure 3 | Response to Anti-BTN3A1 hIgG1 Antibody(mAb1). The H_Vγ9Vδ2(G115) Reporter Jurkat(TCRαβ KO) Cell Line (Cat. GM-C28019) at a concentration of 1E5 cells/well (96-well format) was stimulated with serial dilutions of Anti-BTN3A1 hIgG1 Antibody(mAb1) (Cat. GM-52838AB) in assay buffer (RPMI 1640 + 1% FBS + 1% P.S) for 24 hours. The firefly luciferase activity was measured using the GMOne-Step Luciferase Reporter Gene Assay Kit (Cat. GM-040503). The maximum induction fold was approximately [3.3]. Data are shown by drug mass concentration.



	H_Vγ9Vδ2(G115)(TCR KO) Reporter
EC50	0.3203

Figure 4 | Response to Anti-BTN3A1 hIgG1 Antibody(mAb1). The H_Vγ9Vδ2(G115) Reporter Jurkat(TCRαβ KO) Cell Line (Cat. GM-C28019) at a concentration of 1E5 cells/well (96-well format) was stimulated with serial dilutions of Anti-BTN3A1 hIgG1 Antibody(mAb1) (Cat. GM-52838AB) in assay buffer (RPMI 1640 + 1% FBS + 1% P.S) for 24 hours. The firefly luciferase activity was measured using the GMOne-Step Luciferase Reporter Gene Assay Kit (Cat. GM-040503). The maximum induction fold was approximately [3.3]. Data are shown by drug molar concentration.

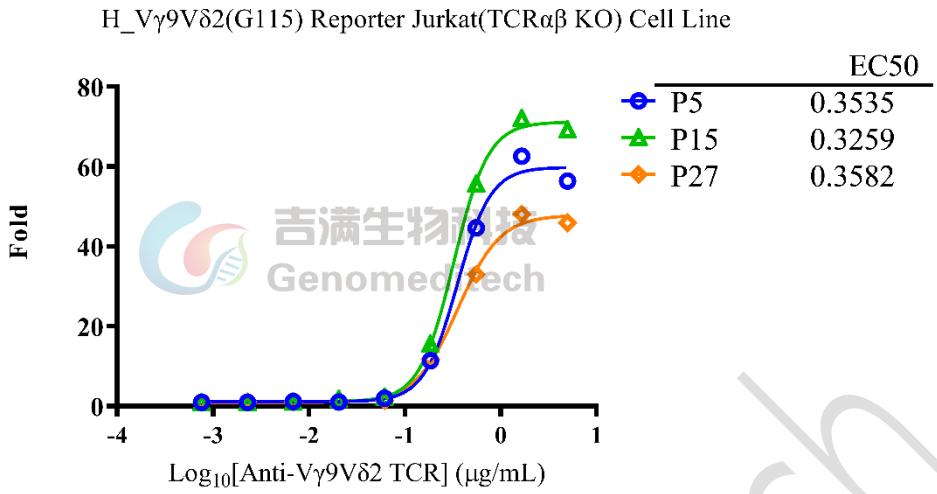


Figure 5 | The passage stability of response to Anti-Vγ9Vδ2 TCR hIgG1 Antibody. The passage 5, 15 and 27 of H_Vγ9Vδ2(G115) Reporter Jurkat(TCRαβ KO) Cell Line (Cat. GM-C28019) at a concentration of 1E5 cells/well in a 96-well format. The wells were coated overnight with serial dilutions of Anti-Vγ9Vδ2 TCR hIgG1 Antibody (Cat. GM-75325AB) in assay buffer (RPMI 1640 + 1% FBS + 1% P.S). After coating, the cells were added and incubated for 24 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.

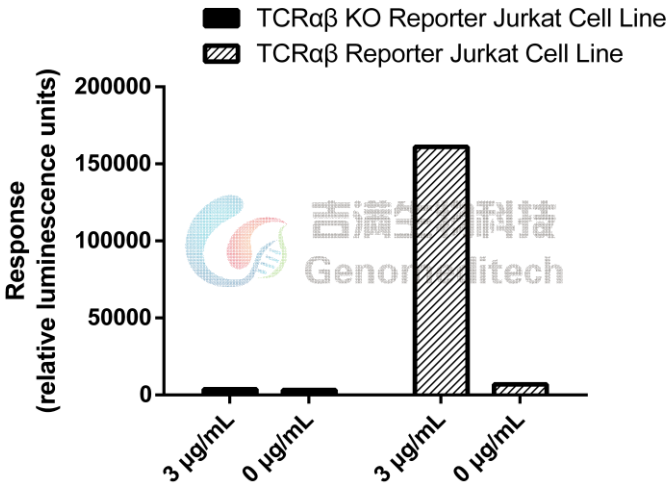
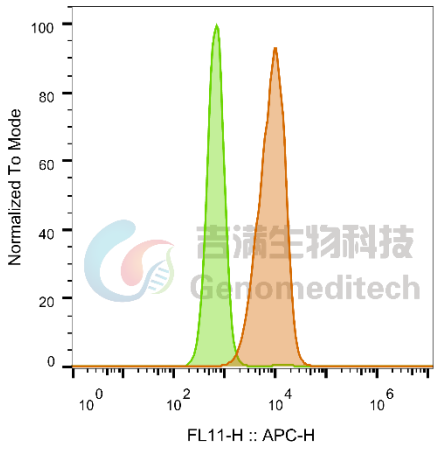


Figure 6 | Response to Anti-CD3 epsilon Antibody [OKT-3 (muromonab)]. The H_Vγ9Vδ2(G115) Reporter Jurkat(TCRαβ KO) Cell Line (Cat. GM-C28019) and TCRαβ Reporter Jurkat Cell Line at a concentration of 1E5 cells/well (96-well format) were stimulated with Anti-CD3 epsilon Antibody [OKT-3 (muromonab)] (Cat. GM-51478AB) concentration of 3 μg/mL or 0 μg/mL in assay buffer (RPMI 1640 + 1% FBS + 1% P.S) for 16 hours. The firefly luciferase activity was measured using the GMOne-Step Luciferase Reporter Gene Assay Kit (Cat. GM-040503).



SampleID	Geometric Mean : FL11-H
Jurkat Anti-BTN3A1+APC-2nd Ab	8236
Jurkat H_IgG+APC-2nd Ab	698

Figure 7 | Jurkat Cell Line was determined by flow cytometry using Anti-BTN3A1 hIgG1 Antibody(mAb1) (Cat. GM-52838AB).

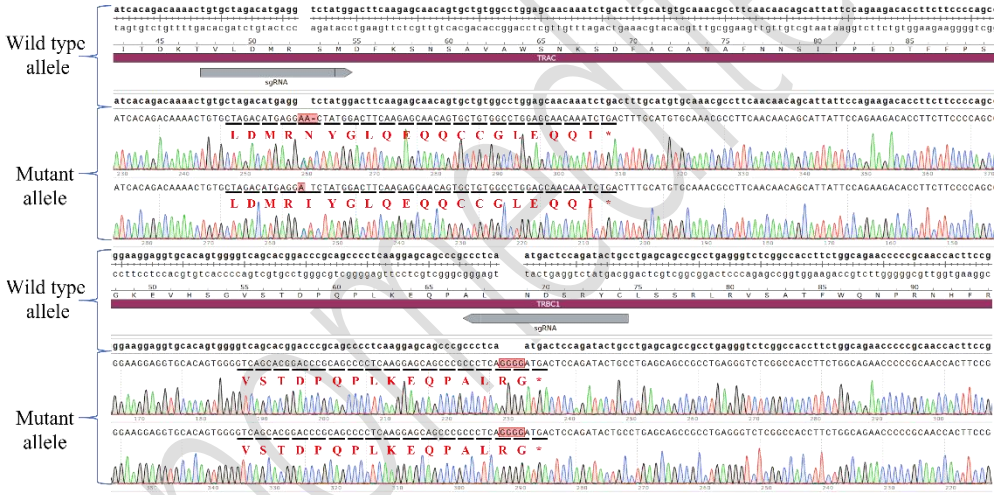


Figure 8 | The Sanger sequencing of the H_Vγ9Vδ2(G115) Reporter Jurkat(TCRαβ KO) Cell Line showed successful knockout of TCRαβ.

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°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).

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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_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: RPMI 1640+10% FBS+1% P.S+3.5 μg/mL Blasticidin+400 μg/mL G418+200 μg/mL Hygromycin+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.

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Related Products

BTN3:Vγ9Vδ2	
TCR Knockout Jurkat Cell Line	H_Vγ9Vδ2(MOP) Reporter Jurkat(TCRαβ KO) Cell Line
H_BTN2A1 HEK-293 Cell Line	H_BTN3A1 HEK-293 Cell Line
Anti-BTN2A1 mIgG1 Antibody(mAb 7.48)	Anti-BTN3A1 hIgG1 Antibody(mAb1)
Anti-H_BTN3A1 hIgG1 Antibody(hu103.2)	Anti-Vγ9Vδ2 TCR hIgG1 Antibody

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