Genomeditech (Shanghai) Co.,Ltd.
Order: +86 021-68455258/50432826/50432825
Toll-free: +86 400 627 9288
Email: service@genomeditech.com

Product Sheet

H_IL17A Reporter 293 DDX35™ Cell Line

Catalog number: GM-C26020

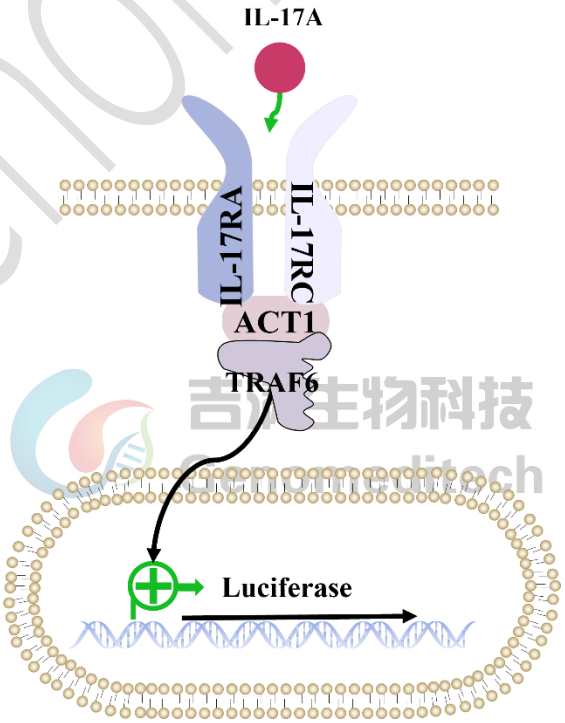
Version 3.3.1.241128

IL-17 is a pro-inflammatory cytokine mainly produced by Th17 cells. It is crucial for immune defense against bacterial and fungal infections and is linked to autoimmune diseases like rheumatoid arthritis, psoriasis, and multiple sclerosis. The IL-17 family includes several members, with IL-17A and IL-17F being the most studied.

The IL-17 signaling pathway starts when IL-17 binds to its receptor, IL-17R, composed of IL-17RA and IL-17RC subunits. This binding activates downstream molecules like ACT1, which then activate the NF-κB and MAPK pathways.

The H_IL17A Reporter 293 DDX35™ Cell Line is a clonal stable cell line constructed using lentiviral technology, constitutive expression of the IL-17RA, IL-17RC and some adapter membrane molecules, along with signal-dependent expression of a luciferase reporter gene. When IL-17A/F binds to IL-17R, it activates downstream signaling pathways, leading to the expression of luciferase. Blockade antibodies can inhibit this signal transmission. 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 IL-17.

The H_IL17A Reporter 293 DDX35™ Cell Line was obtained through extensive monoclonal screening and multiple rounds of monoclonal selection. It possesses high stability, high sensitivity, and high amplification properties, meeting the standards for customers' batch library construction and release experiments.



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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	EMEM(ATCC)+10% FBS+1% P.S
Growth medium	EMEM(ATCC)+10% FBS+1% P.S+3 μg/mL Blasticidin+100 μg/mL Bleomycin+150 μg/mL Hygromycin+1.5 μg/mL Puromycin
Note	Cells should be cultured using ATCC/30-2003 EMEM medium or Growth medium from Genomeditech. The serum should be Cegrogen biotech/A0500-3010 or sourced from Gibco.
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.
EMEM	ATCC/30-2003
Fetal Bovine Serum	Cegrogen biotech/A0500-3010
Pen/Strep	Thermo/15140-122
Blasticidin	Genomeditech/GM-040404
Bleomycin	Genomeditech/GM-040407
Hygromycin	Genomeditech/GM-040403
Puromycin	Genomeditech/GM-040401
Recombinant Human IL-17A (C-6His)	Novoprotein/C774
GMOne-Step Luciferase Reporter Gene Assay Kit	Genomeditech/GM-040503

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Figures

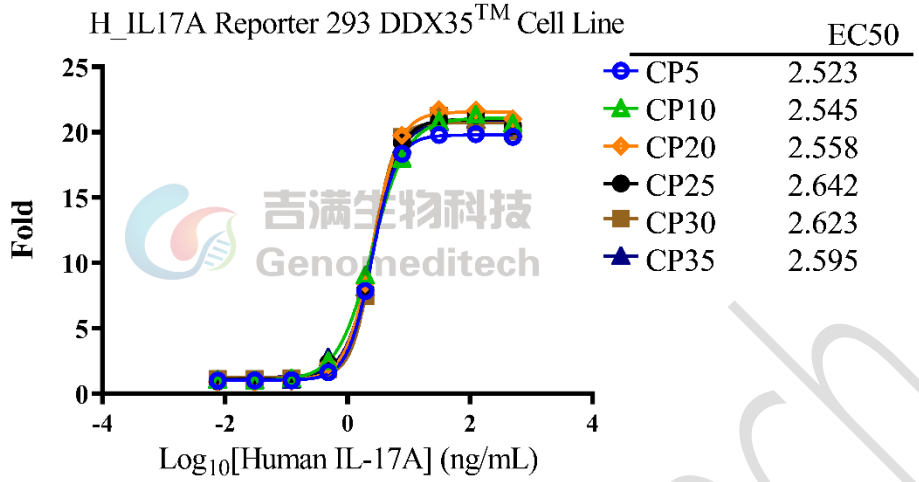

Figure 1 | The passage stability of response to Recombinant Human IL-17A. The passage 5, 10, 20, 25, 30 and 35 of H_IL17A Reporter 293 DDX35™ Cell Line (Cat. GM-C26020) at a concentration of 1.5E4 cells/well (96-well format) was stimulated with serial dilutions of Human IL-17A (Novoprotein/C774) in assay buffer (EMEM(ATCC)+1% FBS+1% P.S) for 7 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.

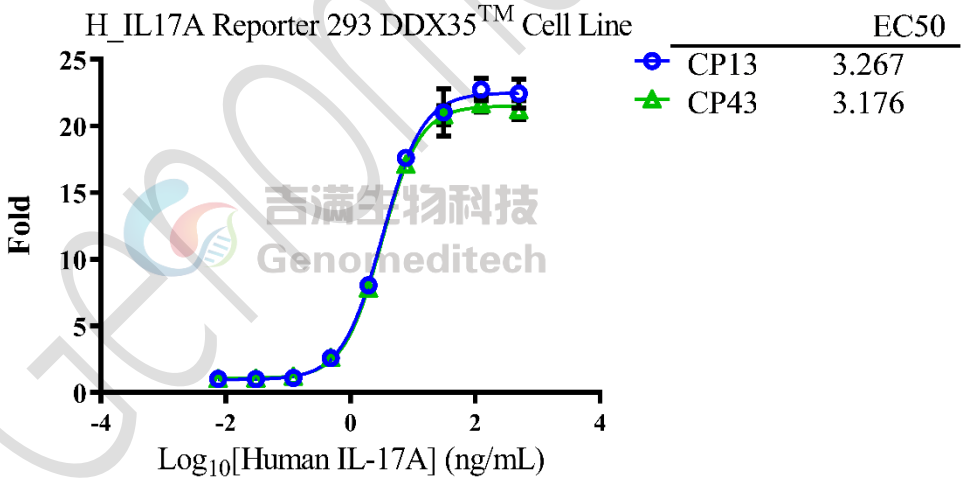

Figure 2 | The passage stability of response to Recombinant Human IL-17A. The passage 13 and 43 of H_IL17A Reporter 293 DDX35™ Cell Line (Cat. GM-C26020) at a concentration of 1.5E4 cells/well (96-well format) was stimulated with serial dilutions of Human IL-17A (Novoprotein/C774) in assay buffer (EMEM(ATCC)+1% FBS+1% P.S) for 7 hours, in triplicate. 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.

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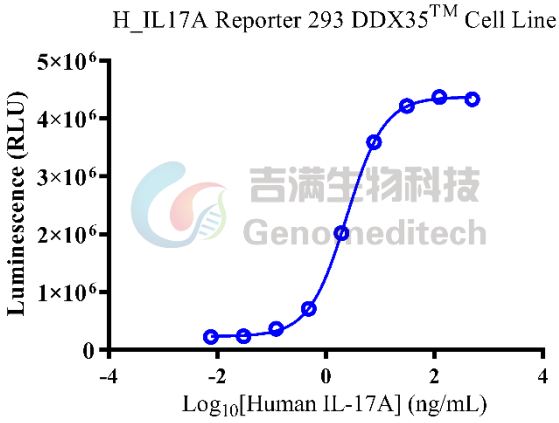
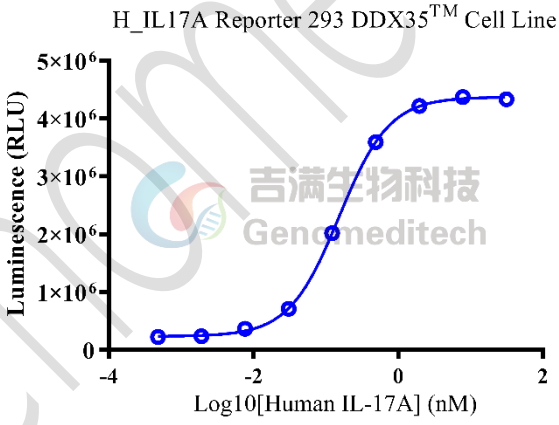


	H_IL17A Reporter 293 DDX35™
EC50	2.436

Figure 3 | Response to Recombinant Human IL-17A (C-6His). The H_IL17A Reporter 293 DDX35™ Cell Line (Cat. GM-C26020) at a concentration of 1.5E4 cells/well (96-well format) was stimulated with serial dilutions of Human IL-17A (C-6His) in assay buffer (EMEM(ATCC)+1% FBS+1% P.S) for 7 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 [23.9]. Data are shown by drug mass concentration.



	H_IL17A Reporter 293 DDX35™
EC50	0.1532

Figure 4 | Response to Recombinant Human IL-17A (C-6His). The H_IL17A Reporter 293 DDX35™ Cell Line (Cat. GM-C26020) at a concentration of 1.5E4 cells/well (96-well format) was stimulated with serial dilutions of Human IL-17A (C-6His) in assay buffer (EMEM(ATCC)+1% FBS+1% P.S) for 7 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 [23.9]. Data are shown by drug molar concentration.

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Cell Recovery

Recovery Medium: EMEM(ATCC)+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: EMEM(ATCC)+10% FBS+1% P.S+3 μg/mL Blasticidin+100 μg/mL Bleomycin+150 μg/mL Hygromycin+1.5 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 30 to 60 seconds 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 1:3 - 1:4 is recommended

Medium Renewal: Every 2 to 3 days

Notes

a) Upon initial revival, a higher number of dead cells and poor adherence are observed, which is normal. Adherence typically recovers within 2 - 3 days. After 2 - 3 passages, the proportion of adherent cells increases, and the cells begin to spread normally.

b) After each passage, there may be 5 - 10% dead cells; however, as the number of passages increases, the recovery rate accelerates, the proportion of dead cells decreases, and the cell growth rate stabilizes.

c) It is recommended to retain cell images after revival and during each observation to assist in assessing cell status. In case of abnormalities, promptly communicate with Genomeditech sales.

Related Products

IL-17	
H_IL17A Reporter 293 Cell Line	
Anti-IL-17A hIgG1 Antibody(Secukinumab)	Anti-IL17A hIgG1 Reference Antibody (Secubio)
IL-23	
H_IL-23 Reporter 293 Cell Line	H_IL-23R HEK-293 Cell Line
TNF:TNFR2:TNFR1	
H_TNFR2 Null Reporter Cell Line	H_TNFR2 Reporter Jurkat Cell Line
H_TNFR2 Reporter V2 Cell Line	Cynomolgus_TNFRSF1B(TNFR2) CHO-K1 Cell Line
H_TNFRSF1B(TNFR2) CHO-K1 Cell Line	H_TNFRSF1B(TNFR2) HEK-293 Cell Line
Membrane Bound H_TNFα CHO-K1 Cell Line	Membrane Bound H_TNFα(cleavage-resistant) CHO-K1 Cell Line
Anti-H_TNFR2 hIgG1 Antibody(1H10)	Anti-H_TNFRSF1B(TNFR2) hIgG1 Antibody(UC2.3.8)
Anti-TNFR1 hIgG1 Antibody(Atrosab)	Anti-TNF-α hIgG1 Antibody（CT-P17）

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