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Product Sheet

H_IL-23 Reporter 293 Cell Line

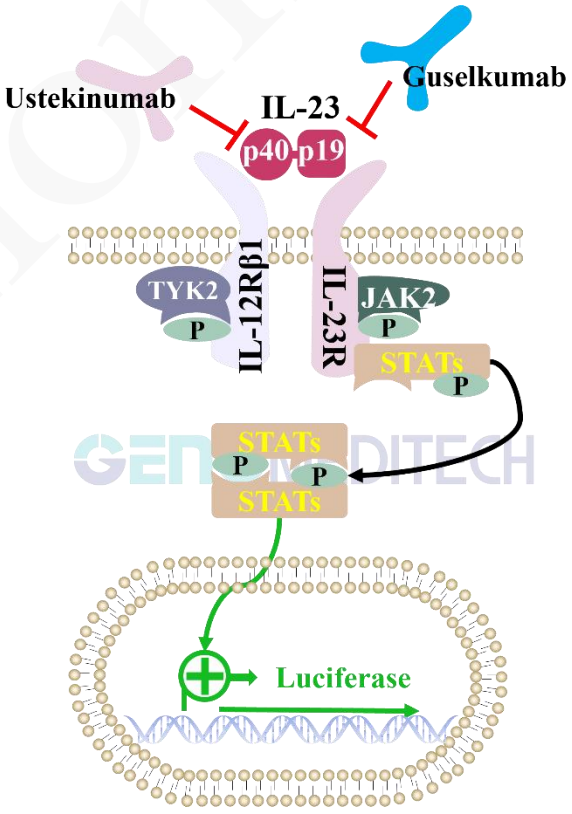
Catalog number: GM-C06722

Version 4.0.26.1030

Interleukin-23 (IL-23) is an inflammatory cytokine that belongs to the IL-12 cytokine family. It consists of two subunits: IL-12B (IL-12p40) and IL-23A (IL-23p19). It's mainly secreted by activated dendritic cells, macrophages, or monocytes. IL-23 is a crucial cytokine for the maintenance and expansion of T helper 17 cells (Th17 cells), enabling them to release their effector cytokines that mediate autoimmune responses.

The functional receptor for IL-23 (IL-23 receptor) is a heterodimer formed between IL-12Rβ1 and IL-23R. IL-23 is associated with autoimmune inflammatory diseases such as colitis, gastritis, psoriasis, and arthritis. Ustekinumab is a monoclonal antibody targeting this cytokine, acting on the P40 subunit, and is used to treat certain autoimmune diseases.

H_IL-23 Reporter 293 Cell Line is a clonal stable HEK-293 cell line constructed using lentiviral technology, constitutive expression of human IL-12Rβ1, IL-23R, along with signal-dependent expression of a luciferase reporter gene. It serves two primary purposes: firstly, when an agonist drug is added, it activates downstream signaling in the reporter gene cells, and luciferase expression is measured to screen or validate agonist drugs targeting IL-12Rβ1 and IL-23R. Secondly, by introducing an IL-23 antagonist antibody to block the signaling activated by IL-23 and measuring luciferase expression, the cell line can be used to screen or validate antagonist drugs targeting IL-23.



Specifications

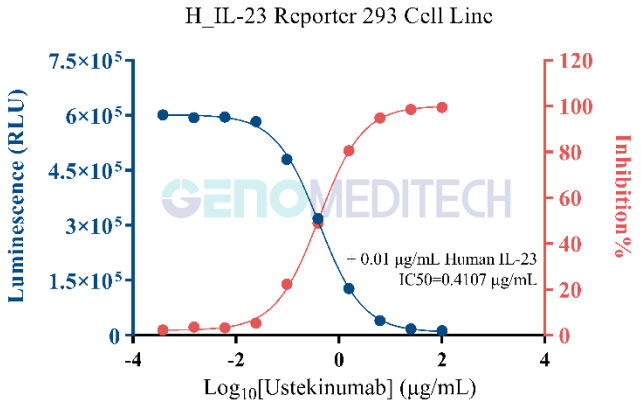
Quantity	5E6 Cells per vial,1 mL
Product Format	1 vial of frozen cells
Shipping	Shipped on dry ice
Storage Conditions	Liquid nitrogen immediately upon receipt
Recovery Medium	DMEM+10% FBS+1% P.S
Growth medium	DMEM+10% FBS+1% P.S+4 μg/mL Blasticidin+125 μg/mL Hygromycin+0.75 μg/mL Puromycin
Note	None
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.
DMEM	Gibco/C11995500BT
Fetal Bovine Serum	ExCell/FSP500
Pen/Strep	Thermo/15140-122
Blasticidin	Genomeditech/GM-040404
Hygromycin	Genomeditech/GM-040403
Puromycin	Genomeditech/GM-040401
Recombinant Human IL-23 (C-6His)	Novoprotein/CJ40
Anti-IL-12/23(p40) hIgG1 Reference Antibody (Ustebio)	Genomeditech/GM-87870MAB
Anti-IL-23R hIgG1 Antibody(5D4)	Genomeditech/GM-87786AB
Anti-Human CD212/IL12RB1 Antibody (148C09)	AtaGenix/FHE37110
Human IL-23A&IL-12B Heterodimer Protein; His Tag	Genomeditech/GM-87924RP
Anti-IL23A hIgG1 Reference Antibody (Gusebio)	Genomeditech/GM-88032MAB
Countess 3 Automated Cell Counter	ThermoFisher/A49891
SpectraMax® L Microplate Luminometer	Moleculardevices/SpectraMax L
96 well round cell culture plate	PakGent/CL-PT096
96-well White Opaque Plate	Thermo/236108
96-well U-bottom Plate	Saining/1014010
GMOne-Step 2.0 Luciferase Reporter Gene Assay Kit	Genomeditech/GM-040513

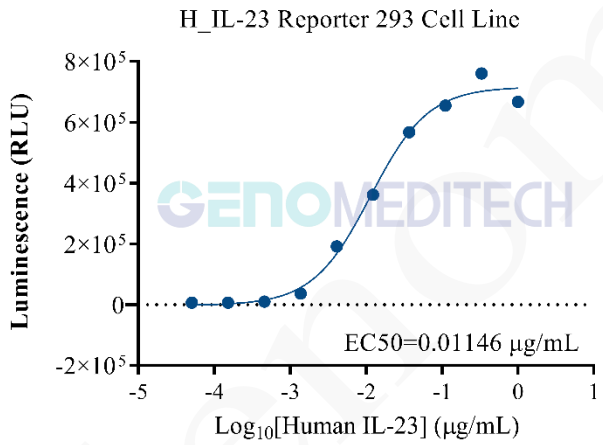
Data

Applications

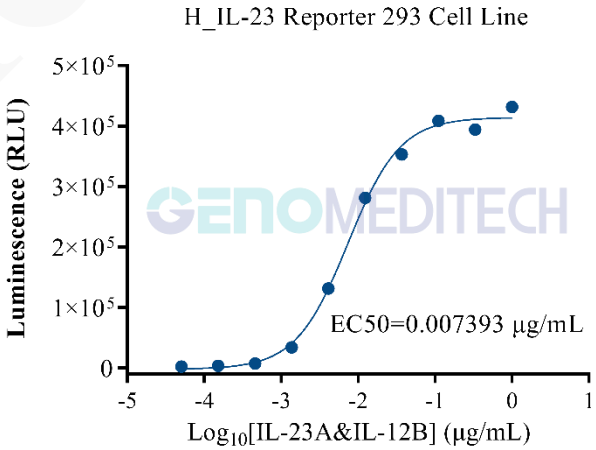


Response to Anti-H_IL-12/23 hIgG1 antibody. Serial dilutions of Anti-IL-12/23(p40) hIgG1 Reference Antibody (Ustebio) (Cat. GM-87870MAB) was incubated with 1 ng/well of Recombinant Human IL-23 (C-6His) (Novoprotein/CJ40) for 1 hour in assay buffer (DMEM + 1% FBS + 1% P.S). After pre-incubation, add the mixture to the H_IL-23 Reporter 293 Cell Line (Cat. GM-C06722) at a density of 1.5E4 cells/well in a 96-well format, and incubate for 16 hours. The firefly luciferase activity was measured using the Luciferase Reporter Assay Kit (Genomeditech). (left Y-axis, relative luminescence units), with inhibition percentages shown on the right Y-axis. Data are shown by drug mass concentration.

Signaling Pathway

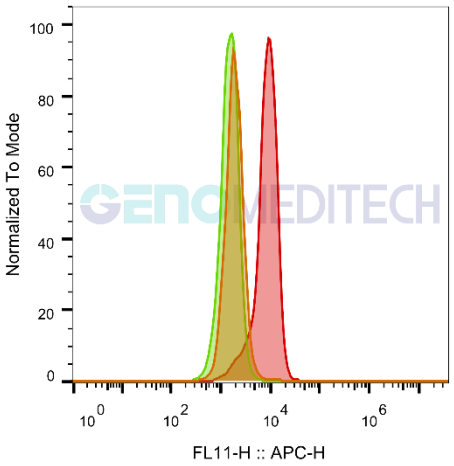


Response to Human IL-23 protein. The H_IL-23 Reporter 293 Cell Line (Cat. GM-C06722) at a concentration of 1.5E4 cells/well (96-well format) was stimulated with serial dilutions of Recombinant Human IL-23 (C-6His) (Novoprotein/CJ40) in assay buffer (DMEM + 1% FBS + 1% P.S) for 16 hours. The firefly luciferase activity was measured using the Luciferase Reporter Assay Kit (Genomeditech). The maximum induction fold was approximately [118.6]. Data are shown by drug mass concentration.



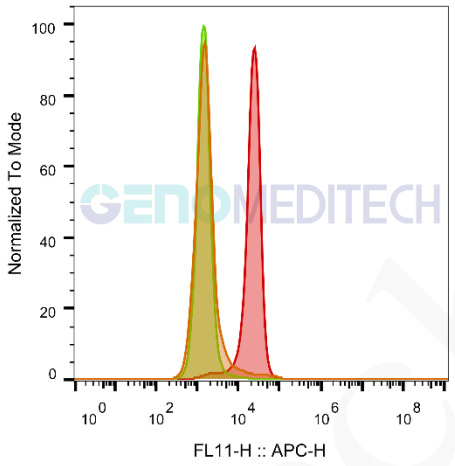
Response to Human IL-23A&IL-12B Heterodimer Protein. The H_IL-23 Reporter 293 Cell Line (Cat. GM-C06722) at a concentration of 1.5E4 cells/well (96-well format) was stimulated with serial dilutions of Human IL-23A&IL-12B Heterodimer Protein (Cat. GM-87924RP) in assay buffer (DMEM + 1% FBS + 1% P.S) for 16 hours. The firefly luciferase activity was measured using the Luciferase Reporter Assay Kit (Genomeditech). The maximum induction fold was approximately [193.8]. Data are shown by drug mass concentration.

Expression



	SampleID	Geometric Mean : FL11-H
	293 anti-IL-23R+APC-2nd Ab	1906
	H_IL-23 Reporter 293 H_IgG+APC-2nd Ab	1517
	H_IL-23 Reporter 293 anti-IL-23R+APC-2nd Ab	8204

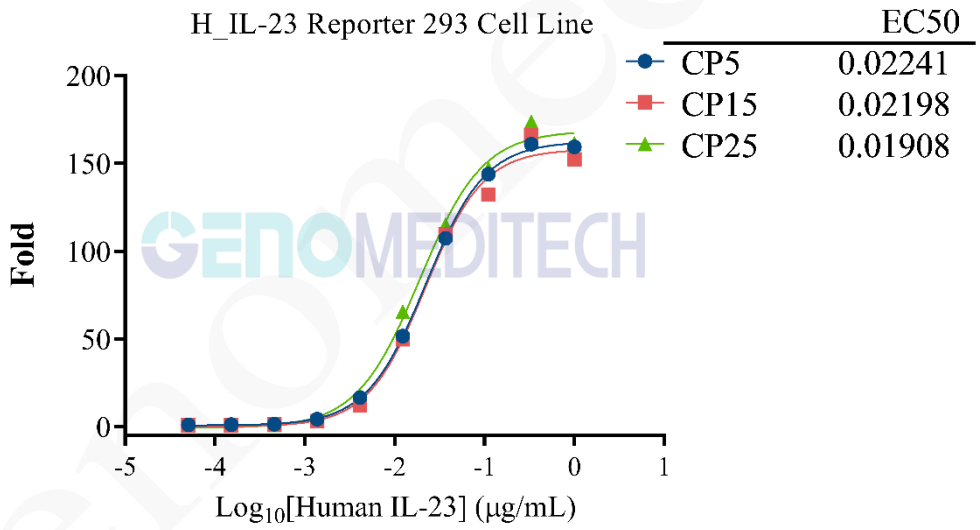
H_IL-23 Reporter 293 Cell Line (Cat. GM-C06722) was determined by flow cytometry using Anti-IL-23R hIgG1 Antibody(5D4) (Cat. GM-87786AB).



	SampleID	Geometric Mean : FL11-H
	293 anti-IL12RB1+APC-2nd Ab	1712
	H_IL-23 Reporter 293 M_IgG+APC-2nd Ab	1474
	H_IL-23 Reporter 293 anti-IL12RB1+APC-2nd Ab	21131

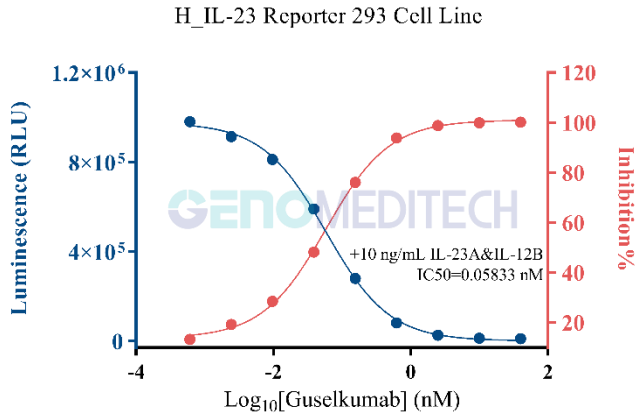
H_IL-23 Reporter 293 Cell Line (Cat. GM-C06722) was determined by flow cytometry using Anti-Human CD212/IL12RB1 Antibody (148C09) (AtaGenix/FHE37110).

Stability



The passage stability of response to Recombinant Human IL-23 (C-6His). The passage 5, 15 and 25 of H_IL-23 Reporter 293 Cell Line (Cat. GM-C06722) at a concentration of 1.5E4 cells/well (96-well format) was stimulated with serial dilutions of Recombinant Human IL-23 (C-6His) (Novoprotein/CJ40) in assay buffer (DMEM + 1% FBS + 1% P.S) for 16 hours. The firefly luciferase activity was measured using the Luciferase Reporter Assay Kit (Genomeditech). Data are shown by drug mass concentration.

Other data



Response to Anti-IL23A hIgG1 Reference Antibody (Gusebio). Serial dilutions of Anti-IL23A hIgG1 Reference Antibody (Gusebio) (Cat. GM-88032MAB) was incubated with 1 ng/well of Recombinant Human IL-23A&IL-12B Heterodimer Protein (Cat. GM-87924RP) for 1 hour in assay buffer (DMEM+1% FBS+1% P.S). After pre-incubation, add the mixture to the H_IL-23 Reporter 293 Cell Line (Cat. GM-C06722) at a density of 1.5E4 cells/well in a 96-well format, and incubate for 7 hours. The firefly luciferase activity was measured using the Luciferase Reporter Assay Kit (Genomeditech). (left Y-axis, relative luminescence units), with inhibition percentages shown on the right Y-axis. Data are shown by drug molar concentration.

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Cell Recovery

Recovery Medium: DMEM+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: DMEM+10% FBS+1% P.S+4 μg/mL Blasticidin+125 μg/mL Hygromycin+0.75 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) Subculturing is necessary when the cell density reaches 80%. It is recommended to perform subculturing at a ratio of 1:3 to 1:4 every 2-3 days. Ensure that the density does not exceed 80%, as overcrowding can lead to reduced viability due to compression.
b) Remove and discard culture medium.
c) Briefly rinse the cell layer with PBS to remove all traces of serum that contains trypsin inhibitor.
d) 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).
e) 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.
f) Add 2.0 mL of growth medium to mix well and aspirate cells by gently pipetting.
g) After centrifugation, resuspend the pellet and add appropriate aliquots of the cell suspension to new culture vessels.
h) 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 thawing, a higher number of dead cells is observed, which is a normal phenomenon. Significant improvement is seen after adaptation. Once the cells reach a stable state, the number of dead cells decreases after subculturing and the cell growth rate becomes stable.

b) Ensure that the cell density does not exceed 80%, as overcrowding may lead to reduced viability due to compression.

Related Products

IL-23	
H_IL-23 Reporter 293 DDX35TM Cell Line	Cynomolgus_IL-23R HEK-293 Cell Line
H_IL-23R HEK-293 Cell Line	H_IL-23R IL12RB1 HEK-293 Cell Line
Membrane bound H_IL-23 CHO-K1 Cell Line	
Anti-IL23A hIgG1 Reference Antibody (Gusebio)	Anti-IL23A hIgG1 Reference Antibody (Risbio)
Anti-IL-23R hIgG1 Antibody(5D4)	
Biotinylated Human IL-23A&IL-12B Heterodimer Protein; His-Avi Tag	Cynomolgus IL-23A & Human IL-12B Heterodimer Protein; His Tag
Cynomolgus IL-23A & Mouse IL-12B Heterodimer Protein; His Tag	Cynomolgus IL-23R Protein; hFc Tag
Human IL-23A & Mouse IL-12B Heterodimer Protein; His Tag	Human IL-23A&IL-12B Heterodimer Protein; His Tag
Human IL-23R Protein; hFc Tag	Human IL-23R Protein; His Tag
Mouse IL-23A&IL-12B Heterodimer Protein; His Tag	
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_TNFR1 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_TNFRSF1B(TNFR2) hIgG1 Antibody(UC2.3.8)	Anti-TNFR1 hIgG1 Antibody(Atrosab)
Anti-TNF-α hIgG1 Reference Antibody (Adabio)	
Cynomolgus TNF alpha Protein; His Tag	Cynomolgus TNFR2 Protein; His Tag
Human TNF alpha Protein; hFc Tag	Human TNF alpha Protein; His Tag
Human TNFR1 Protein; hFc Tag	Human TNFR1 Protein; His Tag
Human TNFR2 Protein; hFc Tag	Human TNFR2 Protein; His Tag
TL1A:DR3(TNFRSF25)	
H_TNFRSF25(DR3) Reporter Jurkat Cell Line	H_TNFSF15(TL1A) Reporter Cell Line
Mouse_TNFRSF25(DR3) Reporter Jurkat Cell Line	Cynomolgus_TNFRSF25(DR3) CHO-K1 Cell Line
Cynomolgus_TNFSF15(TL1A) HEK-293 Cell Line	H_TNFRSF25(DR3) CHO-K1 Cell Line
H_TNFRSF25(DR3) HEK-293 Cell Line	H_TNFSF15(TL1A) CHO-K1 Cell Line
H_TNFSF15(TL1A) HEK-293 Cell Line	Mouse_TNFSF15(TL1A) HEK-293 Cell Line
Anti-H_TNFSF15(TL1A) hIgG1 Antibody(PF-06480605)	Anti-H_TNFSF15(TL1A) hIgG1 Antibody(Tulisokibart、PRA-023)
Anti-H_TNFSF15(TL1A) hIgG4 Antibody	Anti-TL1A hIgG1 Reference Antibody (Duvbio)

Anti-TL1A hIgG1 Reference Antibody (Tulbio)	Anti-TNFRSF25(DR3) hIgG1 Antibody(PTX-35)
Biotinylated Cynomolgus TL1A Protein; His-Avi Tag	Biotinylated Human TL1A Protein; His-Avi Tag
Cynomolgus TL1A Protein; His Tag	Human TL1A Protein; hFc Tag
Human TL1A Protein; His Tag	Human TL1A Protein; His Tag (Trimer Design)
ITGA4 ITGB7	
Cynomolgus_α4β7 CHO-K1 Cell Line	H_ITGB7 CHO-K1 Cell Line
H_ITGB7 HEK-293 Cell Line	H_MADCAM1 CHO-K1 Cell Line
H_MADCAM1 HEK-293 Cell Line	H_α4β7 CHO-K1 Cell Line
H_α4β7 HEK-293 Cell Line	H_αEβ7 CHO-K1 Cell Line
H_αEβ7 HEK-293 Cell Line	H_αEβ7 K562 Cell Line
Anti-Integrin α4β7 hIgG1 Reference Antibody (Vedobio)	
Human MAdCAM-1 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:

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