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Product Sheet

H_TLR8 Reporter 293 Cell Line

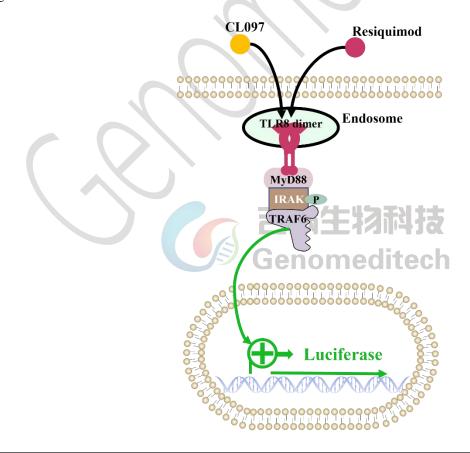
Catalog number: GM-C15427

Version 3.3.1.241226

TLR8 (Toll-Like Receptor 8) is a pattern recognition receptor on the cell membrane, part of the Toll-like receptor family. It is primarily found in immune cells like monocytes and dendritic cells, recognizing viral RNA and pathogen-associated patterns. TLR8 activation boosts immune responses, including cytokine production and immune cell activation, playing a crucial role in antiviral immunity and inflammation.

TLR8 signaling is mediated by its intracellular TIR (Toll/IL-1 receptor) domain. Upon ligand binding, TLR8 recruits adaptor proteins (e.g., MyD88) and activates downstream pathways like NF-κB and MAPK. This leads to cytokine production, such as IL-6 and TNF-α, enhancing the immune response. TLR8 also improves the host's resistance to viral infections by regulating antiviral gene expression, making it vital for immune homeostasis and pathogen response.

H_TLR8 Reporter 293 Cell Line is a clonal stable HEK-293 cell line constructed using lentiviral technology, constitutive expression of the TLR8 gene, along with signal-dependent expression of a luciferase reporter gene. When Resiquimod binds to TLR8, it activates downstream signaling pathways, leading to the expression of luciferase. 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 TLR8.





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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+1.5 µg/mL Puromycin

Cells should be cultured using ATCC/30-2003 EMEM medium or Growth medium from **Note**

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% CO₂

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
Puromycin	Genomeditech/GM-040401
Resiquimod	MCE/HY-13740
CL097	InvivoGen/tlrl-c97
GMOne-Step Luciferase Reporter Gene Assay Kit	Genomeditech/GM-040503

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Figures

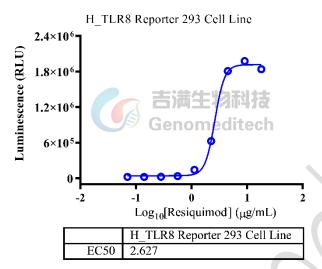


Figure 1 | Response to Resiquimod. The H_TLR8 Reporter 293 Cell Line (Cat. GM-C15427) at a concentration of 2E4 cells/well (96-well format) was stimulated with serial dilutions of Resiquimod (MCE/HY-13740) in assay buffer (EMEM(ATCC) + 1% FBS + 1% P.S) for 7 hours. The firefly luciferase activity was measured using the GMOne-Step Luciferase Reporter Gene Assay Kit (Cat. GM-040503). The maximum induction fold was approximately [75.5]. Data are shown by drug mass concentration.

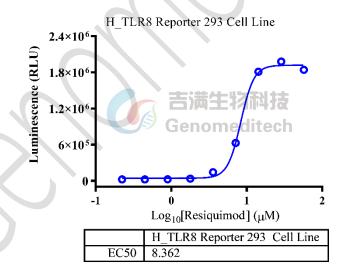


Figure 2 | Response to Resiquimod. The H_TLR8 Reporter 293 Cell Line (Cat. GM-C15427) at a concentration of 2E4 cells/well (96-well format) was stimulated with serial dilutions of Resiquimod (MCE/HY-13740) in assay buffer (EMEM(ATCC) + 1% FBS + 1% P.S) for 7 hours. The firefly luciferase activity was measured using the GMOne-Step Luciferase Reporter Gene Assay Kit (Cat. GM-040503). The maximum induction fold was approximately [75.5]. Data are shown by drug molar concentration.



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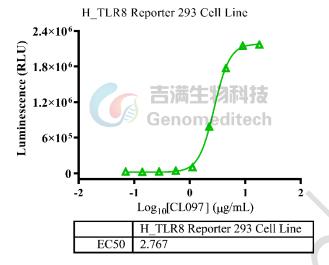


Figure 3 | Response to CL097. The H_TLR8 Reporter 293 Cell Line (Cat. GM-C15427) at a concentration of 2E4 cells/well (96-well format) was stimulated with serial dilutions of CL097 (InvivoGen/tlrl-c97) in assay buffer (EMEM(ATCC) + 1% FBS + 1% P.S) for 7 hours. The firefly luciferase activity was measured using the GMOne-Step Luciferase Reporter Gene Assay Kit (Cat. GM-040503). The maximum induction fold was approximately [79.4]. Data are shown by drug mass concentration.

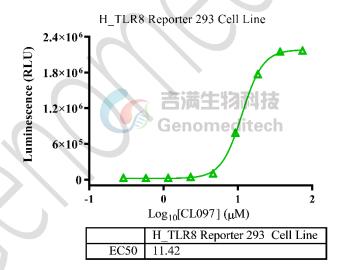


Figure 4 | Response to CL097. The H_TLR8 Reporter 293 Cell Line (Cat. GM-C15427) at a concentration of 2E4 cells/well (96-well format) was stimulated with serial dilutions of CL097 (InvivoGen/tlrl-c97) in assay buffer (EMEM(ATCC) + 1% FBS + 1% P.S) for 7 hours. The firefly luciferase activity was measured using the GMOne-Step Luciferase Reporter Gene Assay Kit (Cat. GM-040503). The maximum induction fold was approximately [79.4]. 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% CO₂ 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+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

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Medium Renewal: Every 2 to 3 days

Notes

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.

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

TLR7 Mouse_TLR7 Reporter 293 Cell Line H_TLR7 Reporter 293 Cell Line

TLR9

Mouse_TLR9 Reporter 293 Cell Line H_TLR9 Reporter 293 Cell Line

TLR8

H TLR8 HEK-293 Cell Line

STING

H STING KO U937 Cell Line H_STING KO THP1 Cell Line

STING KO Reporter THP1 Cell Line STING Reporter HEK-293 Cell Line

STING Reporter THP1 Cell Line STING Reporter U937 Cell Line

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