Genomeditech (Shanghai) Co.,Ltd.
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Product Sheet

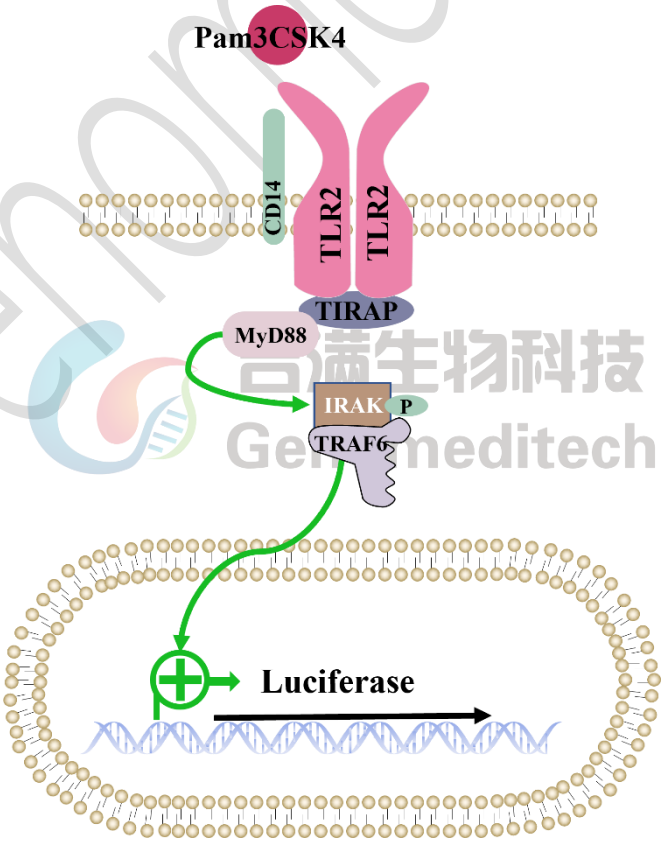
H_TLR2 Reporter 293 Cell Line

Catalog number: GM-C15493

Version 3.3.1.250703

TLR2 (Toll-like receptor 2) is a widely distributed transmembrane protein belonging to the Toll-like receptor family and acts as an important pattern recognition receptor (PRR) in the innate immune system. TLR2 is mainly expressed on the surface of immune cells such as monocytes, macrophages, and dendritic cells, and is capable of recognizing various pathogen-associated molecular patterns (PAMPs), such as bacterial lipoproteins, peptidoglycan, and lipopolysaccharides. By binding to its ligands, TLR2 can activate downstream signaling pathways, induce the production of inflammatory cytokines, and promote the initiation and regulation of immune responses. Studies have shown that TLR2 plays a key role not only in defending against infections and clearing pathogens, but is also closely associated with a variety of inflammatory diseases, autoimmune disorders, and tumorigenesis.

H_TLR2 Reporter 293 Cell Line is a clonal stable 293 cell line constructed using lentiviral technology, constitutive expression of the TLR2 gene, along with signal-dependent expression of a luciferase reporter gene. When Pam3CSK4 binds to TLR2, 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 TLR2.



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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+400 μg/mL G418+1.5 μg/mL Puromycin
Note	Cells should be cultured using ATCC/30-2003 EMEM medium or Growth medium from Genomeditech.
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	ExCell/FSP500
Pen/Strep	Thermo/15140-122
Blasticidin	Genomeditech/GM-040404
G418	Genomeditech/GM-040402
Puromycin	Genomeditech/GM-040401
Pam3CSK4 TFA	MCE/HY-P1180A
GMOne-Step 2.0 Luciferase Reporter Gene Assay Kit	Genomeditech/GM-040513

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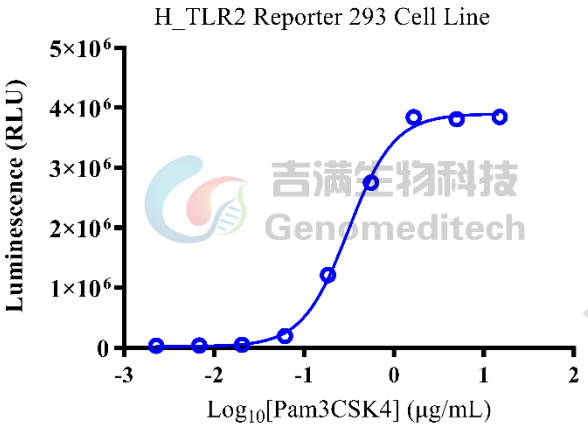
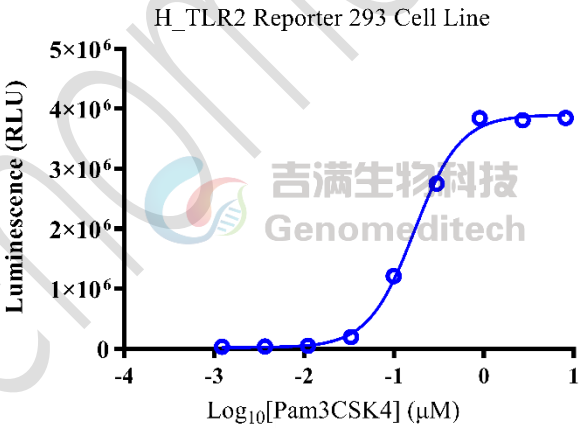
Figures



	H_TLR2 Reporter 293 Cell Line
EC50	0.3126

Figure 1 | Response to Pam3CSK4(TFA). The H_TLR2 Reporter 293 Cell Line (Cat. GM-C15493) at a concentration of 2E4 cells/well (96-well format) was stimulated with serial dilutions of Pam3CSK4(TFA) (MCE/HY-P1180A) in assay buffer (ATCC EMEM+1% FBS+1% P.S) for 6 hours. The firefly luciferase activity was measured using the GMOne-Step 2.0 Luciferase Reporter Gene Assay Kit (Cat. GM-040513C). The maximum induction fold was approximately [119.5]. Data are shown by drug mass concentration.



	H_TLR2 Reporter 293 Cell Line
EC50	0.1688

Figure 2 | Response to Pam3CSK4(TFA). The H_TLR2 Reporter 293 Cell Line (Cat. GM-C15493) at a concentration of 2E4 cells/well (96-well format) was stimulated with serial dilutions of Pam3CSK4(TFA) (MCE/HY-P1180A) in assay buffer (ATCC EMEM+1% FBS+1% P.S) for 6 hours. The firefly luciferase activity was measured using the GMOne-Step 2.0 Luciferase Reporter Gene Assay Kit (Cat. GM-040513C). The maximum induction fold was approximately [119.5]. 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+400 μg/mL G418+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

TLR7	
H_TLR7 Reporter 293 Cell Line	Mouse_TLR7 Reporter 293 Cell Line
TLR9	
H_TLR9 Reporter 293 Cell Line	Mouse_TLR9 Reporter 293 Cell Line
TLR8	
H_TLR8 Reporter 293 Cell Line	H_TLR8 HEK-293 Cell Line
STING	
H_STING KO THP1 Cell Line	H_STING KO U937 Cell Line
STING KO Reporter THP1 Cell Line	STING Reporter HEK-293 Cell Line
STING Reporter THP1 Cell Line	STING Reporter U937 Cell Line

License Agreement:

By purchasing and using this cell line product, the user voluntarily agrees to accept and abide by the following policies:

- This cell line product is restricted to research use only and shall not be used for any commercial purposes.
- This product is strictly prohibited from being used in the diagnosis or treatment of human or animal diseases, and shall not be directly used in experiments involving humans.
- Users are not permitted to modify the cell line in any way, nor to share, distribute, sell, sublicense, or otherwise transfer the licensed materials or their progeny to other laboratories, departments, research institutes, hospitals, universities, biotechnology companies, or any other third parties, except for research activities outsourced on behalf of the licensee.
- If the product is intended to be transferred to a third party, used for commercial development, preclinical or clinical drug functional validation, commercial production testing, or any other applications beyond the scope of this statement, prior

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