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Product Sheet

H_PRL(prolactin) Reporter Cell Line

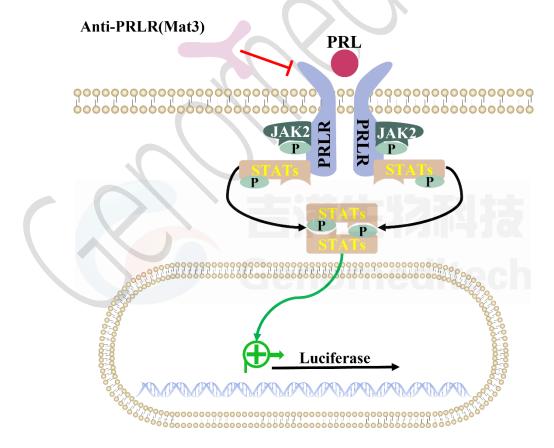
Catalog number: GM-C19941

Version 3.3.1.241226

PRL (Prolactin) is a hormone from the anterior pituitary that promotes mammary gland development and milk secretion. It also regulates reproduction, immunity, and metabolism, with secretion influenced by factors like TRH and dopamine.

The PRL signaling pathway is mediated by the PRL receptor (PRLR), which activates JAK2 and subsequently STAT5, leading to target gene expression. This pathway is crucial for mammary cell proliferation and differentiation and impacts other tissues' functions. Abnormalities in this pathway may be linked to diseases like breast cancer and infertility.

H_PRL(prolactin) Reporter Cell Line is a clonal stable cell line constructed using lentiviral technology, constitutive expression of the PRLR gene, along with signal-dependent expression of a luciferase reporter gene. When PRL binds to PRLR, 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 PRL.





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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 DMEM+10% FBS+1% P.S

Growth medium DMEM+10% FBS+1% P.S+4 μg/mL Blasticidin+0.75 μg/mL Puromycin

Note None

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.
DMEM	VivaCell/C3110-0500
Fetal Bovine Serum	Cegrogen biotech/A0500-3010
Pen/Strep	Thermo/15140-122
Blasticidin	Genomeditech/GM-040404
Puromycin	Genomeditech/GM-040401
Recombinant Human Prolactin Protein	Sino Biological/10275-H08B
Anti-PRLR hIgG1 Antibody(Mat3)	Genomeditech/GM-27198AB
GMOne-Step Luciferase Reporter Gene Assay Kit	Genomeditech/GM-040503



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Figures



Figure 1 | Response to Recombinant Human Prolactin Protein. The H_PRL(prolactin) Reporter Cell Line (Cat. GM-C19941) at a concentration of 1.5E4 cells/well (96-well format) was stimulated with serial dilutions of Recombinant Human Prolactin Protein (Sino Biological/10275-H08B) in assay buffer (DMEM + 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). The maximum induction fold was approximately [201.2]. Data are shown by drug mass concentration.

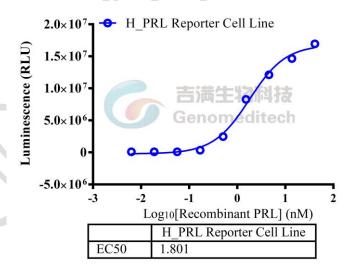


Figure 2 | Response to Recombinant Human Prolactin Protein. The H_PRL(prolactin) Reporter Cell Line (Cat. GM-C19941) at a concentration of 1.5E4 cells/well (96-well format) was stimulated with serial dilutions of Recombinant Human Prolactin Protein (Sino Biological/10275-H08B) in assay buffer (DMEM + 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). The maximum induction fold was approximately [201.2]. Data are shown by drug molar concentration.



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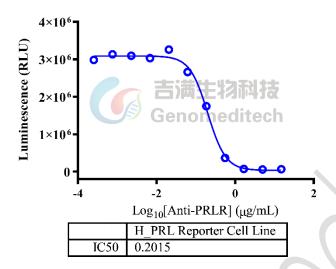


Figure 3 | Response to Anti-PRLR hIgG1 Antibody(Mat3). Serial dilutions of the Anti-PRLR hIgG1 Antibody(Mat3) (Cat. GM-27198AB) was incubated with 1.5E4 cells/well of the H_PRL(prolactin) Reporter Cell Line (Cat. GM-C19941) in a 96-well plate for 1 hour in assay buffer (DMEM + 1% FBS + 1% P.S). Subsequently, the Recombinant Human Prolactin Protein (Sino Biological/10275-H08B) at a concentration of 21 ng/well was added, and the coculture proceeded for an additional 15 hours. Firefly luciferase activity is then measured using the GMOne-Step Luciferase Reporter Gene Assay Kit (Cat. GM-040503). The results indicated maximum blocking folds of approximately [46.3]. Data are shown by drug mass concentration.

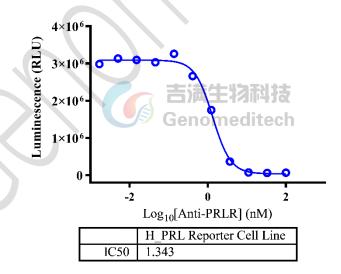


Figure 4 | Response to Anti-PRLR hIgG1 Antibody(Mat3). Serial dilutions of the Anti-PRLR hIgG1 Antibody(Mat3) (Cat. GM-27198AB) was incubated with 1.5E4 cells/well of the H_PRL(prolactin) Reporter Cell Line (Cat. GM-C19941) in a 96-well plate for 1 hour in assay buffer (DMEM + 1% FBS + 1% P.S). Subsequently, the Recombinant Human Prolactin Protein (Sino Biological/10275-H08B) at a concentration of 21 ng/well was added, and the coculture proceeded for an additional 15 hours. Firefly luciferase activity is then measured using the GMOne-Step

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Luciferase Reporter Gene Assay Kit (Cat. GM-040503). The results indicated maximum blocking folds of approximately [46.3]. Data are shown by drug molar concentration.

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% 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: DMEM+10% FBS+1% P.S+4 µg/mL Blasticidin+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) 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.



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- 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 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.

Related Products

PRL:PRLR	
Cynomolgus_PRLR CHO-K1 Cell Line	H_PRLR CHO-K1 Cell Line
Mouse_PRLR CHO-K1 Cell Line	Mouse_PRLR HEK-293 Cell Line
Anti-PRLR hIgG1 Antibody(Mat3)	Anti-PRLR hIgG1 Reference Antibody(Mat3)

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