Human PTH1R (Luc) HEK293 Reporter Cell Data Sheet

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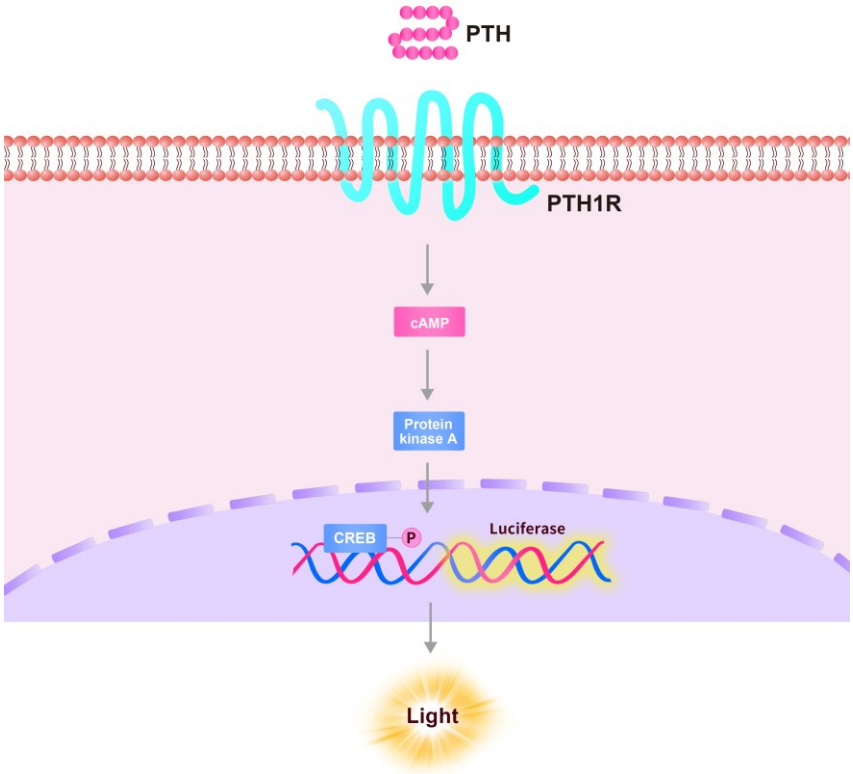
Catalog No.	Size
CHEK-ATF194	2 × (1 vial contains ~5×10^6 cells)

• *Description*

The Human PTH1R (Luc) HEK293 Reporter Cell was engineered to not only express CREB signaling response element, but also express the receptor full length human PTH1R (Gene ID:5745). which can drive luciferase expressing systems by PTH1R agonists or PTH stimulation. In the absence of agonist or PTH, the PTH1R receptor is not activated and luminescence signal is low. In the presence of agonist or PTH, the PTH1R pathway-activated luminescence can be detected in a dose-dependent manner.

• *Application*

- Screen for agonists that can bind and activate PTH1R



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• *Cell Line Profile*

Cell line	Human PTH1R (Luc) HEK293 Reporter Cell
Host Cell	HEK293
Property	Adherent
Complete Growth Medium	DMEM + 10% FBS
Selection Marker	Puromycin (2 μg/mL) + Hygromycin (20 μg/mL)
Incubation	37°C with 5% CO_2
Doubling Time	22-24 hours
Transduction Technique	Lentivirus

• *Materials Required for Cell Culture*

- DMEM medium (Gibco, Cat. No. 11965-092)
- Fetal bovine serum (CellMax, Cat. No. SA211.02)
- Puromycin (InvivoGen, Cat. No. ant-pr-5b)
- Hygromycin B (Invitrogen, Cat. No. 10687010)
- 0.25% Trypsin-EDTA (1X), Phenol Red (Gibco, Cat. No. 25200-056)
- Penicillin-Streptomycin (Gibco, Cat. No. 15140-122)
- Phosphate Buffered Saline (1X) (HyClone, Cat. No. SH30256.01)
- Complete Growth Medium: DMEM + 10% FBS, 1% PS
- Culture Medium: DMEM + 10% FBS, Puromycin (2 μg/mL) + Hygromycin (20 μg/mL), 1% PS
- Freeze Medium: 90% FBS, 10% (V/V) DMSO
- T-75 Culture flask (Corning, 430641)
- Cryogenic storage vials (SARSTEDT, 72.379.007)
- Thermostat water bath
- Centrifuge
- Luna cell counter (Logos Biosystems, LUNA-Ⅱ)
- CO_2 Incubator (Thermo, 3111)
- Biological Safety Cabinet (Thermo, 1389)

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• *Recovery*

1. Thaw the vial by gentle agitation in a 37°C water bath. To reduce the possibility of contamination, keep the cap out of the water. Thawing should be rapid (approximately 2 minutes).
2. Remove the vial from the water bath as soon as the contents are thawed, and decontaminate by spraying with 70% ethanol. All the operations from this point on should be carried out under strict aseptic conditions.
3. Transfer the vial contents to a centrifuge tube containing 4.0 mL complete growth medium and spin at approximately 1000 rpm for 5 minutes.
4. Resuspend cell pellet with 5 mL complete growth medium and transfer the cell suspension into T-75 flask containing 10-15 mL of pre-warmed complete growth medium.
5. Incubate at 37°C with 5% CO_2 incubator until the cells are ready to be split.

• *Subculture*

1. Remove and discard culture medium.
2. Wash the cells once with sterile PBS.
3. Add 2 mL of 0.25% trypsin to cell culture flask. Place the flask at 37°C for 2-3 minutes, until 90% of the cells have detached.
4. Add 6.0 to 8.0 mL of culture medium and aspirate cells by gently pipetting.
5. Add appropriate aliquots of the cell suspension to new culture vessel.
6. Incubate at 37°C with 5% CO_2 incubator.

Subcultivation Ratio: A subcultivation ratio of 1:6 to 1:10 is recommended.

Medium Renewal: Every 2 to 3 days.

Note: After recovery for 1-2 generations with the complete growth medium not containing the selection marker, if the cell state is well, changing to the culture medium containing the selection marker.

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• *Cryopreservation*

1. Remove and discard spent medium.
2. Detach cells from the cell culture flasks with 0.25% trypsin.
3. Centrifuge at 1000 rpm for 5 min at RT to pellet cells.
4. Resuspend the cell pellets with complete growth medium and count viable cells.
5. Centrifuge at 1000 rpm for 5 min at RT and resuspend cells in freezing medium to a concentration of 5×10^6 to 1×10^7 cells/mL.
6. Aliquot into cryogenic storage vials. Place vials in a programmable cooler or an insulated box placed in a –80°C freezer overnight, then transferring to liquid nitrogen storage.

• *Storage*

- **Product format:** Frozen
- **Storage conditions:** Liquid nitrogen immediately upon receipt

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• *Application*

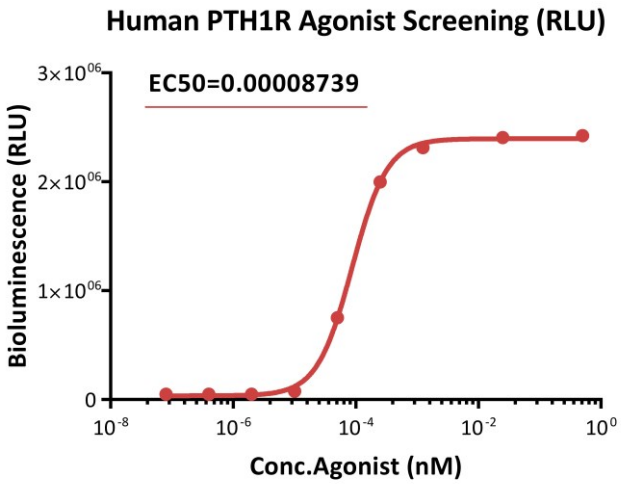

Fig1. Bioactivity analysis of human PTH1R agonist (RLU). This reporter cell was incubated with serial dilutions of human PTH1R agonist. The EC50 of human PTH1R agonist (Teriparatide) was approximately 0.00008739 nM.

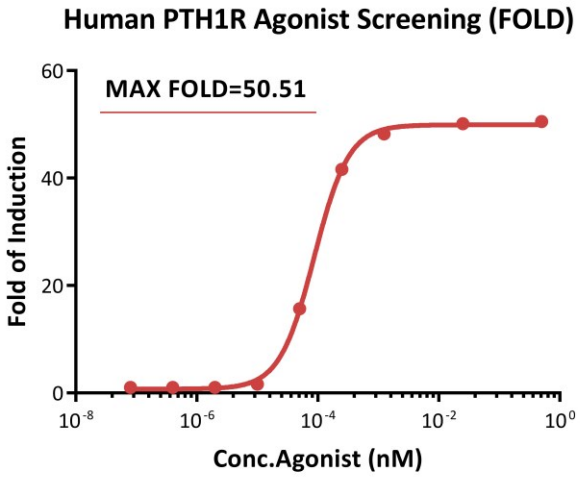

Fig2. Bioactivity analysis of human PTH1R agonist (FOLD). This reporter cell was incubated with serial dilutions of human PTH1R agonist. The max induction fold of human PTH1R agonist (Teriparatide) was approximately 50.51.

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• *License Disclosure*

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• *Related Products*

Products	**Cat.No.**
Human GCGR (Luc) HEK293 Reporter Cell	CHEK-ATF103
Human GIPR (Luc) HEK293 Reporter Cell	CHEK-ATF104
Human THRA (Luc) HEK293 Reporter Cell	CHEK-ATF180
Human THRB (Luc) HEK293 Reporter Cell	CHEK-ATF181
Human GLP-2R (Luc) HEK293 Reporter Cell	CHEK-ATF128
Human GLP-1R (Luc) HEK293 Reporter Cell	CHEK-ATF096