

# Human THRB (Luc) HEK293 Reporter Cell Data Sheet

## Human THRB (Luc) HEK293 Reporter Cell

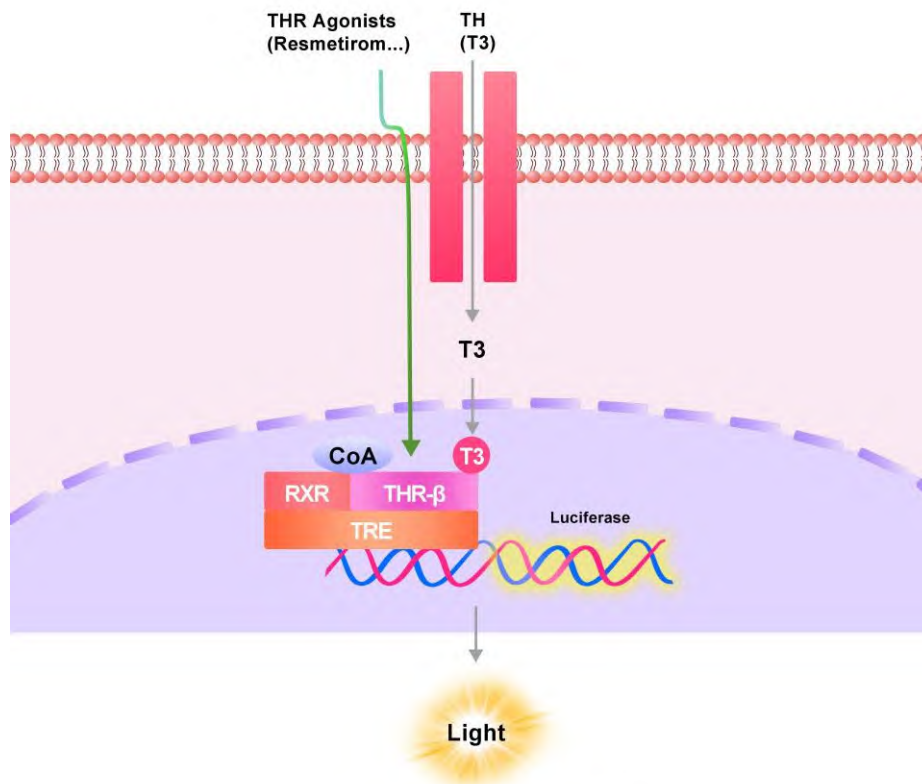
| Catalog No. | Size   |
|-------------|--|
| CHEK-ATF181 | 2 × (1 vial contains ~5×10 <sup>6</sup> cells) |

### • Description

The Human THRB (Luc) HEK293 Reporter Cell was engineered to not only express thyroid hormone response element (TRE), but also express the receptor full length human THRB (Gene ID: 7068), which can drive luciferase expressing systems by thyroid hormone (TH) or thyroid hormone receptor (THR) agonists stimulation. In the absence of TH or THR agonists, the THRB receptor is not activated and luminescence signal is low. In the presence of TH or THR agonists, the THRB pathway-activated luminescence can be detected in a dose-dependent manner.

### • Application

- Screen for THR-β-selective agonists.



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## • Cell Line Profile

|                               |   |
|-------------------------------|---|
| <b>Cell line</b>              | Human THRB (Luc) HEK293 Reporter Cell       |
| <b>Host Cell</b>              | HEK293                                      |
| <b>Property</b>               | Adherent                                    |
| <b>Complete Growth Medium</b> | DMEM + 10% FBS                              |
| <b>Selection Marker</b>       | Puromycin (2 µg/mL) + Hygromycin (20 µg/mL) |
| <b>Incubation</b>             | 37°C with 5% CO <sub>2</sub>                |
| <b>Doubling Time</b>          | 22-24 hours                                 |
| <b>Transduction Technique</b> | 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
- Culture Medium: DMEM + 10% FBS, Puromycin (2 µg/mL), Hygromycin (20 µg/mL)
- 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<sub>2</sub> 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<sub>2</sub> 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<sub>2</sub> incubator.

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

**Medium Renewal:** Every 2 to 3 days.

# Human THRβ (Luc) HEK293 Reporter Cell Data Sheet

## • *Cryopreservation*

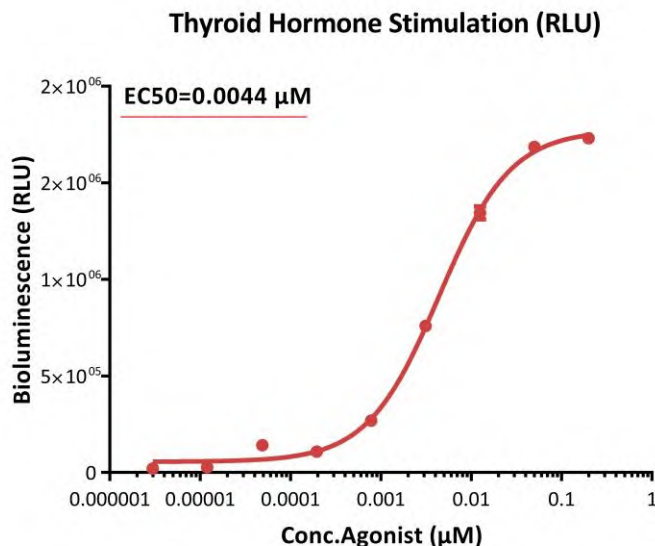
1. Remove and discard spent medium.
2. Detach cells from the cell culture flasks with 0.25% trypsin.
3. Centrifuge at 1000 rpm for 5 min at RT to pellet cells.
4. Resuspend the cell pellets with complete growth medium and count viable cells.
5. Centrifuge at 1000 rpm for 5 min at RT and resuspend cells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

## • *Storage*

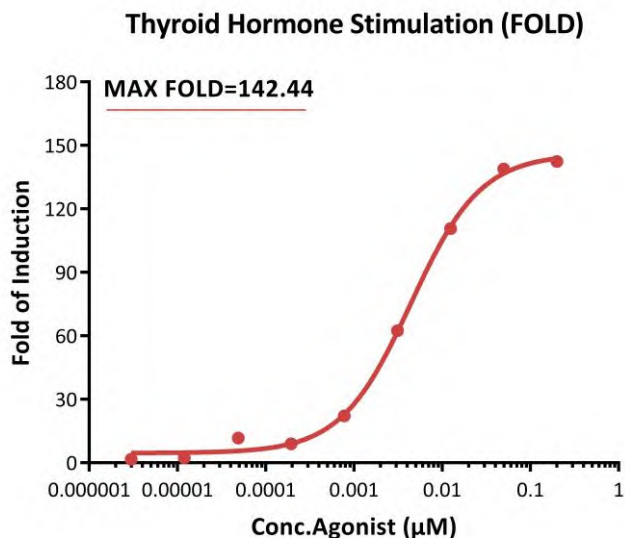
- **Product format:** Frozen
- **Storage conditions:** Liquid nitrogen immediately upon receipt

# Human THRβ (Luc) HEK293 Reporter Cell Data Sheet

• *Signaling Bioassay*



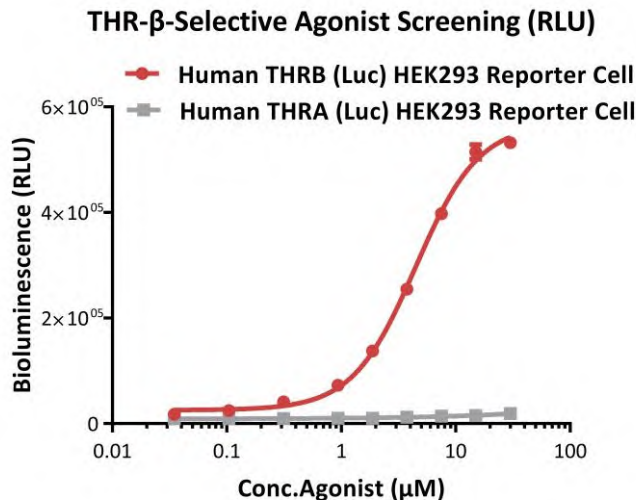
**Fig1. Response to thyroid hormone (RLU).** This reporter cell was incubated with serial dilutions of Liothyronine (a dual THR- $\alpha$  and THR- $\beta$  agonist). The EC50 of Liothyronine was approximately 0.0044  $\mu$ M.



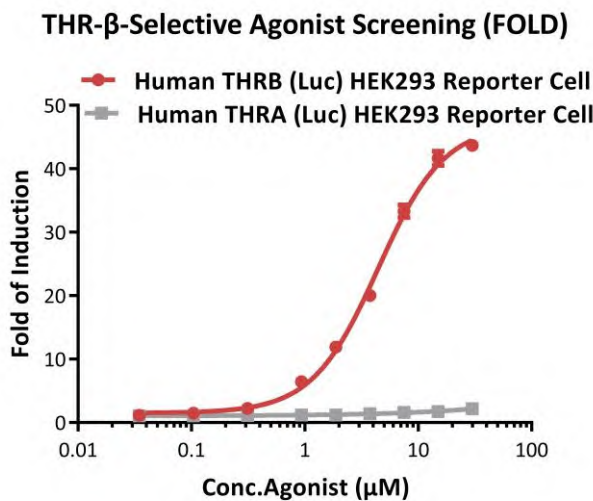
**Fig2. Response to thyroid hormone (FOLD).** This reporter cell was incubated with serial dilutions of Liothyronine (a dual THR- $\alpha$  and THR- $\beta$  agonist). The max induction fold was approximately 142.44.

# Human THRB (Luc) HEK293 Reporter Cell Data Sheet

• *Application*



**Fig3. Bioactivity analysis of THR-β-selective agonist (RLU).** The Human THRB (Luc) HEK293 Reporter Cell and Human THRA (Luc) HEK293 Reporter Cell (Cat.No.CHEK-ATF180) were incubated with serial dilutions of Resmetirom (a THR-β-selective agonist), respectively. The EC50 of Resmetirom determined on Human THRB (Luc) HEK293 Reporter Cell was approximately 4.4 µM.



**Fig4. Bioactivity analysis of THR-β-selective agonist (FOLD).** The Human THRB (Luc) HEK293 Reporter Cell and Human THRA (Luc) HEK293 Reporter Cell (Cat.No.CHEK-ATF180) were incubated with serial dilutions of Resmetirom (a THR-β-selective agonist), respectively. The max induction fold of Resmetirom determined on Human THRB (Luc) HEK293 Reporter Cell was approximately 43.7, and on Human THRA (Luc) HEK293 Reporter Cell was approximately 2.2, which exhibited higher potency and selectivity for THR-β over THR-α.

# Human THRB (Luc) HEK293 Reporter Cell Data Sheet

• *License Disclosure*

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• *Related Products*

**Products**

**Cat.No.**

Human THRA (Luc) HEK293 Reporter Cell

CHEK-ATF180