

# Human TSLP R (Luc) HEK293 Reporter Cell Data Sheet

## Human TSLP R (Luc) HEK293 Reporter Cell

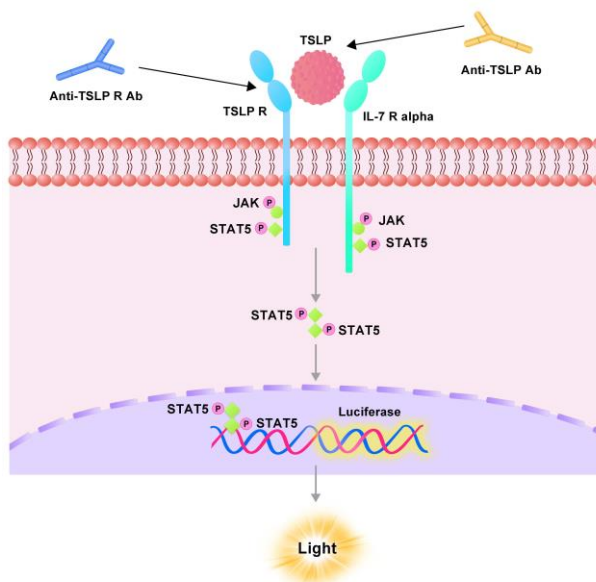
Catalog No.	Size
CHEK-ATF045	2 × (1 vial contains ~5×10 <sup>6</sup> cells)

### • Description

The Human TSLP R (Luc) HEK293 Reporter Cell was engineered to not only express STAT5 signaling response element, but also express the receptors full length human TSLP R (Gene ID: 64109) and IL-7 R alpha (Gene ID: 3575). When stimulated with human TSLP protein, the TSLP/TSLP R interaction drives STAT5-mediated luminescence. Inhibition of TSLP binding to TSLP R by either anti-TSLP or anti-TSLP R antibodies results in a decrease in luminescence.

### • Application

- Screen for anti-human TSLP or anti-human TSLP R neutralizing antibody.



### • Cell Line Profile

Cell line	Human TSLP R (Luc) HEK293 Reporter Cell
Host Cell	HEK293
Property	Adherent
Complete Growth Medium	DMEM + 10% FBS
Selection Marker	Puromycin (2 µg/mL) + Zeocin (100 µg/mL)
Incubation	37°C with 5% CO <sub>2</sub>
Doubling Time	22-24 hours
Transduction Technique	Lentivirus

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## • *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)
- Zeocin (Invitrogen, Cat.No.R25001)
- Complete Growth Medium: DMEM + 10% FBS
- Culture Medium: DMEM + 10% FBS, Puromycin (2 µg/mL), Zeocin (100 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)

## • *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.

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## • *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.

## • *Cryopreservation*

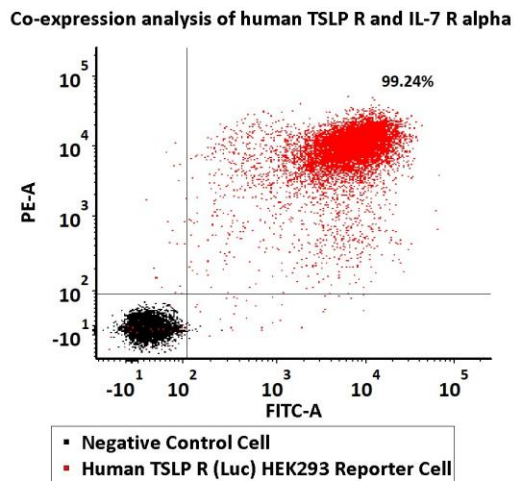
1. Remove and discard spent medium.
2. Detach cells from the cell culture flasks with 0.25% trypsin.
3. Centrifuge at 1000 rpm for 5 min at RT to pellet cells.
4. Resuspend the cell pellets with complete growth medium and count viable cells.
5. Centrifuge at 1000 rpm for 5 min at RT and resuspend cells 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°C freezer overnight, then transferring to liquid nitrogen storage.

## • *Storage*

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

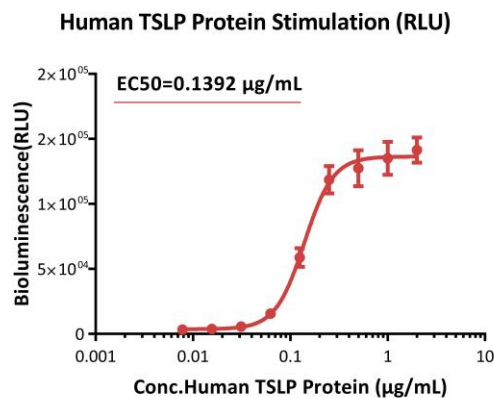
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## • Receptor Assay



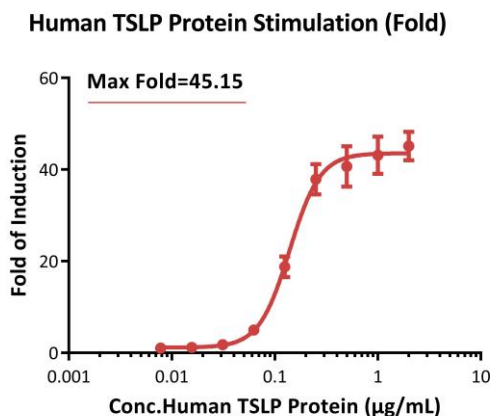
**Fig1. Co-expression analysis of human TSLP R and IL-7 R alpha on Human TSLP R (Luc) HEK293 Reporter Cell by FACS.** Cell surface staining was performed on Human TSLP R (Luc) HEK293 Reporter Cell or negative control cell using PE-labeled anti-TSLP R antibody and FITC-labeled anti-IL-7 R alpha antibody.

## • Signaling Bioassay



**Fig2. Response to human TSLP protein (RLU).** The Human TSLP R (Luc) HEK293 Reporter Cell was stimulated with serial dilutions of human TSLP protein (Cat.No.TSP-H52Hb). The EC50 was approximately 0.1392 µg/mL.

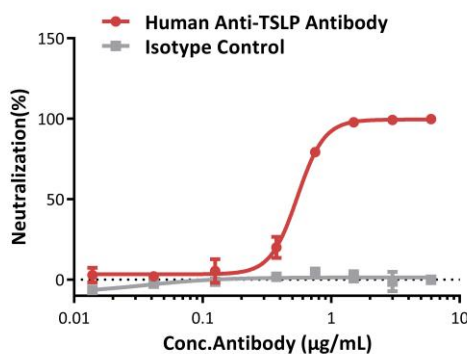
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**Fig3. Response to human TSLP protein (Fold).** The Human TSLP R (Luc) HEK293 Reporter Cell was stimulated with serial dilutions of human TSLP protein (Cat.No.TSP-H52Hb). The max induction fold was approximately 45.15.

• **Application**

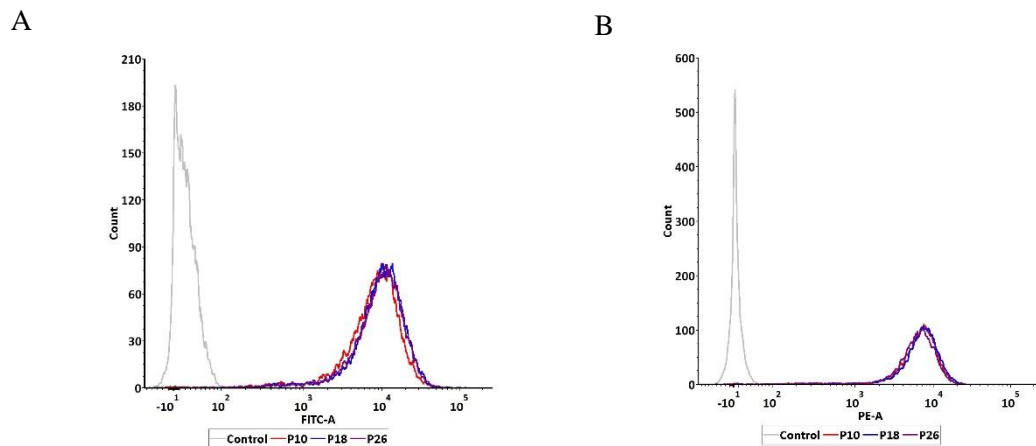
**Anti-human TSLP Neutralizing Antibody Screening**



**Fig4. Inhibition of human TSLP protein-induced reporter activity by anti-human TSLP neutralizing antibody.** This reporter cell was incubated with serial dilutions of antibodies in the presence of human TSLP protein (Cat.No.TSP-H52Hb) with a final concentration of 0.3 µg/mL. The EC50 of anti-human TSLP neutralizing antibody is approximately 0.55 µg/mL.

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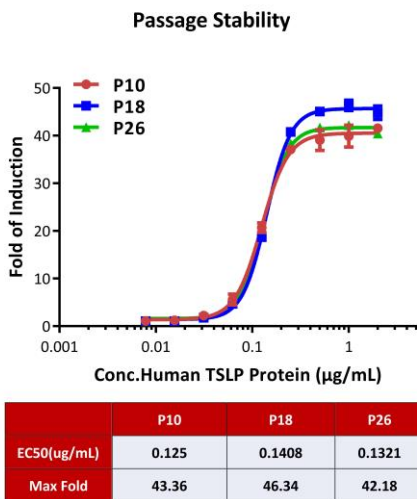
## • Passage Stability



Passage	MFI for IL-7 R alpha (FITC)	MFI for TSLP R (PE)
P10	8267.26	6540.34
P18	9690.88	6838.46
P26	9584.24	6113.65

**Fig5. Passage stability analysis of receptors expression by FACS.** Flow cytometry surface staining of human TSLP R and IL-7 R alpha on Human TSLP R (Luc) HEK293 Reporter Cell demonstrates consistent mean fluorescent intensity across across passage10-26. (A) Human IL-7 R alpha expression analysis. (B) Human TSLP R expression analysis.

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**Fig6. Passage stability analysis by Signaling Bioassay.** The continuously growing Human TSLP R (Luc) HEK293 Reporter Cell was stimulated with serial dilutions of human TSLP protein. Human TSLP protein stimulated response demonstrates passage stabilization (fold induction and EC50) across passage 10-26.

## • License Disclosure

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## • Related Products

### Products

Human TSLP Protein

Human IL-5 R alpha/CD131 (Luc) HEK293 Reporter Cell

Human IL-4 R alpha/IL-13 R alpha 1 (Luc) HEK293 Reporter Cell

### Cat.No.

TSP-H52Hb

CHEK-ATF074

CHEK-ATF075