

Human IL-17 RA/IL-17 RC (Luc) HEK293 Reporter Cell

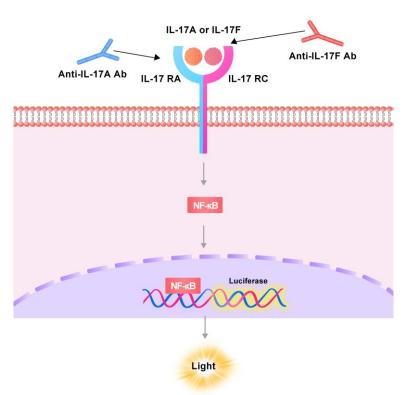
Catalog No.	Size
CHEK-ATF133	$2 \times (1 \text{ vial contains} \sim 5 \times 10^6 \text{ cells})$

• Description

The Human IL-17 RA/IL-17 RC (Luc) HEK293 Reporter Cell was engineered to express NF-kB signaling response element driving luciferase expressing systems. When stimulated with human IL-17A or human IL-17F protein, receptor-mediated signaling can drive NF-kB-mediated luminescence. Neutralization of biological effect of human IL-17A or human IL-17F protein by corresponding antibody results in a decrease in luminescence.

• Application

• Screen for neutralizing antibodies blocking the stimulation of human IL-17A or human IL-17F protein.



Human IL-17 RA/IL-17 RC (Luc) HEK293 Reporter Cell



• Cell Line Profile

Cell line	Human IL-17 RA/IL-17 RC (Luc) HEK293 Reporter Cell	
Host Cell	HEK293	
Property	Adherent	
Complete Growth Medium	DMEM + 10% FBS	
Selection Marker	Puromycin (2 μg/mL) + Zeocin (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)
- Zeocin (Invitrogen, Cat.No.R25001)
- Complete Growth Medium: DMEM + 10% FBS
- Culture Medium: DMEM + 10% FBS, Puromycin (2 μg/mL), Zeocin (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)
- CO₂ 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% CO₂ 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% CO₂ 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 5×10^6 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



• Signaling Bioassay



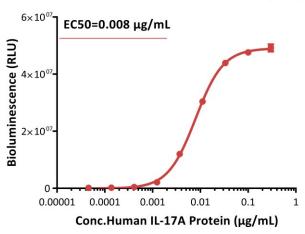


Fig1. Response to human IL-17A protein (RLU). The Human IL-17 RA/IL-17 RC (Luc) HEK293 Reporter Cell was stimulated with serial dilutions of human IL-17A protein (Cat.No.ILA-H5219). The EC50 was approximately 0.008 μg/mL.

Human IL-17A Protein Stimulation (FOLD)

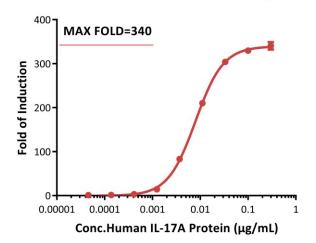


Fig2. Response to human IL-17A protein (FOLD). The Human IL-17 RA/IL-17 RC (Luc) HEK293 Reporter Cell was stimulated with serial dilutions of human IL-17A protein (Cat.No.ILA-H5219). The max induction fold was approximately 340.



Human IL-17F Protein Stimulation (RLU)

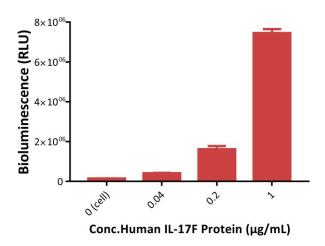


Fig3. Response to human IL-17F protein (RLU). The Human IL-17 RA/IL-17 RC (Luc) HEK293 Reporter Cell was stimulated with serial dilutions of human IL-17F protein (Cat.No.ILF-H5244). The max induction fold was approximately 52.82.

Human IL-17F Protein Stimulation (FOLD)

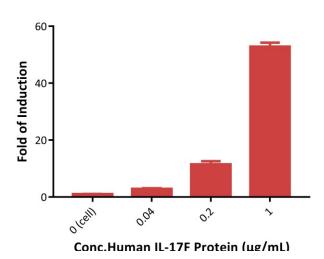


Fig4. Response to human IL-17F protein (FOLD). The Human IL-17 RA/IL-17 RC (Luc) HEK293 Reporter Cell was stimulated with serial dilutions of human IL-17F protein (Cat.No.ILF-H5244). The max induction fold was approximately 52.82.



• Application

Anti-human IL-17A Neutralizing Antibody Screening

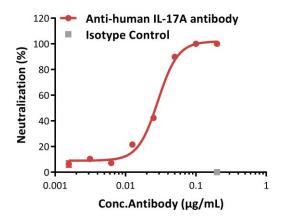


Fig5. Inhibition of human IL-17A protein-induced reporter activity by anti-human IL-17A neutralizing antibody. This reporter cell was incubated with serial dilutions of antibodies in the presence of human IL-17A protein (Cat.No.ILA-H5219) with a final concentration of 0.01 μg/mL. The EC50 of anti-human IL-17A neutralizing antibody (Bimekizumab) is approximately 0.02893 μg/mL.

Anti-human IL-17F Neutralizing Antibody Screening

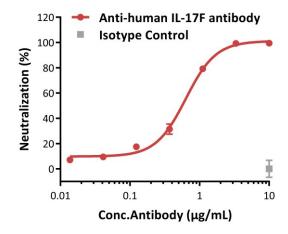


Fig6. Inhibition of human IL-17F protein-induced reporter activity by anti-human IL-17F neutralizing antibody. This reporter cell was incubated with serial dilutions of antibodies in the presence of human IL-17F protein (Cat.No.ILF-H5244) with a final concentration of $0.5 \mu g/mL$. The EC50 of anti-human IL-17F neutralizing antibody (Bimekizumab) is approximately $0.6346 \mu g/mL$.



Passage Stability

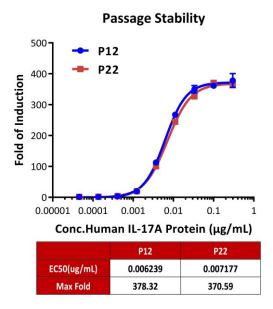


Fig7. Passage stability analysis by Signaling Bioassay. The continuously growing Human IL-17 RA/IL-17 RC (Luc) HEK293 Reporter Cell was stimulated with serial dilutions of human IL-17A protein. Human IL-17A protein stimulated response demonstrates passage stabilization (fold induction and EC50) across passage 12-22.

• License Disclosure

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Related Products

<u>Products</u>	<u>Cat.No.</u>
Human IL17A Protein, premium grade	ILA-H5219
Human IL17F Protein, His Tag	ILF-H5244