Human HVEM (Luc) HEK293 Reporter Cell Data Sheet

Human HVEM (Luc) HEK293 Reporter Cell

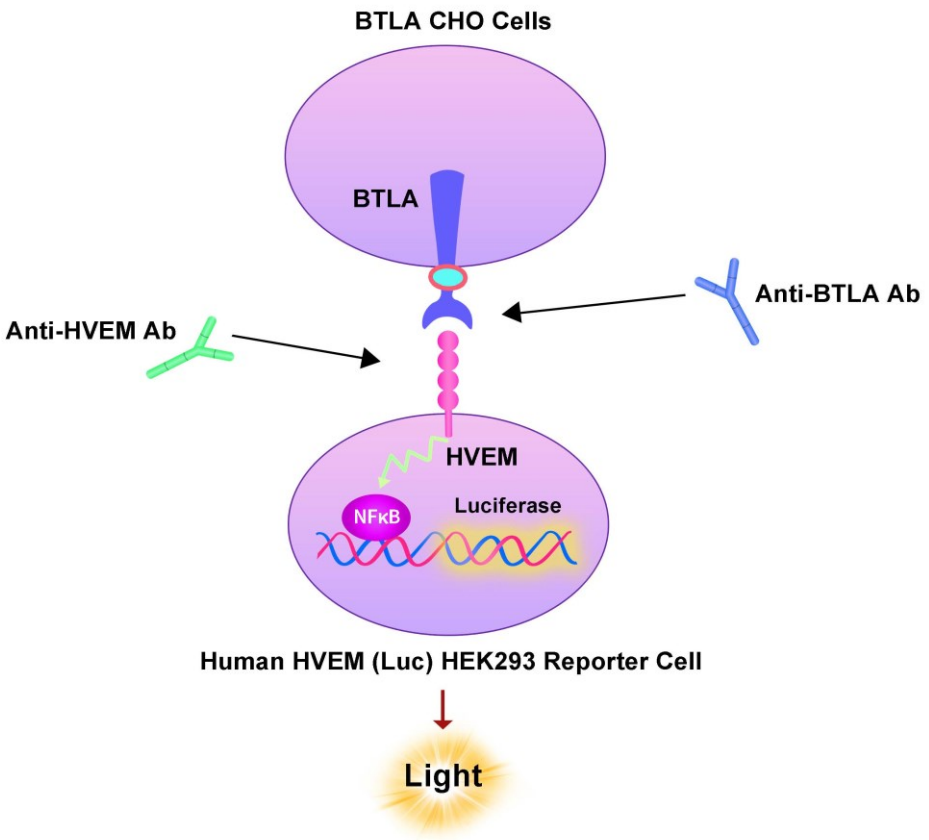
Catalog No.	Size
CHEK-ATF105	2 × (1 vial contains ~5×10^6 cells)

• *Description*

The Human HVEM (Luc) HEK293 Reporter Cell was engineered to not only express NF-kB signaling response element, but also express the receptor full length human HVEM (Gene ID: 8764). When co-cultured with target cells expressing human BTLA, the BTLA/HVEM interaction drives NF-kB-mediated luminescence. Blocking the BTLA/HVEM interaction by either anti-BTLA or anti-HVEM antibodies results in a decrease in luminescence.

• *Application*

- Screen for anti-human BTLA antibody or anti-human HVEM antibody blocking BTLA/HVEM interaction.



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• *Cell Line Profile*

Cell line	Human HVEM (Luc) HEK293 Reporter Cell
Host Cell	HEK293
Property	Adherent
Complete Growth Medium	DMEM + 10% FBS
Selection Marker	Hygromycin (20 μg/mL)
Incubation	37℃ 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)
- Complete Growth Medium: DMEM + 10% FBS
- Culture Medium: DMEM + 10% FBS, 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_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℃ 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℃ 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.

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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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• ***Receptor Assay***

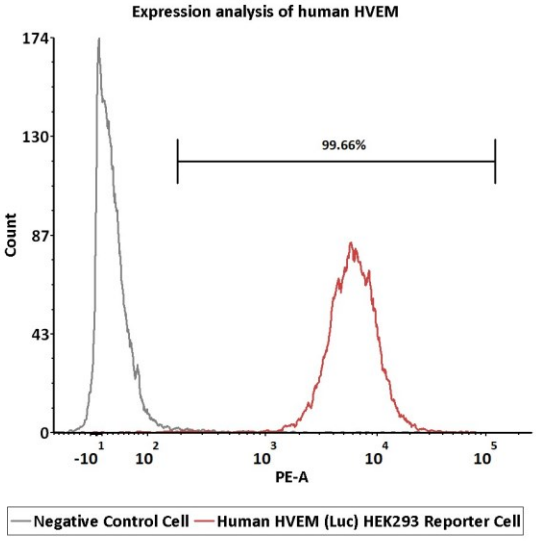

Fig1. Expression analysis of human HVEM on Human HVEM (Luc) HEK293 Reporter Cell by FACS. Cell surface staining was performed on Human HVEM (Luc) HEK293 Reporter Cell or negative control cell using PE-labeled anti-human HVEM antibody.

• ***Signaling Bioassay***

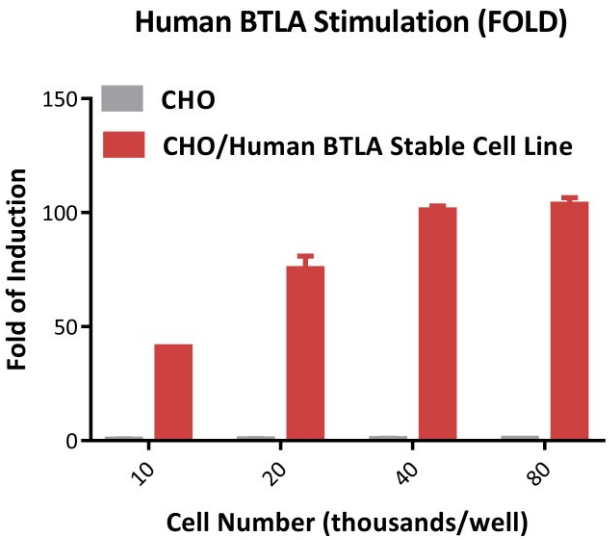

Fig2. Response to human BTLA (FOLD). This reporter cell was incubated with CHO/Human BTLA Stable Cell Line (Cat.No.SCCHO-ATP110) at four different cell densities. The human BTLA overexpressing on CHO cells can activate HVEM signaling with the max induction fold 103.69 at the density of $8x10^4$ cells/well.

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• *Application*

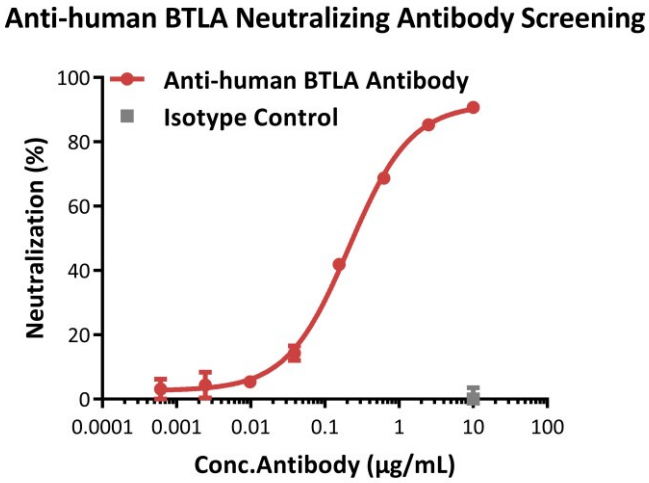
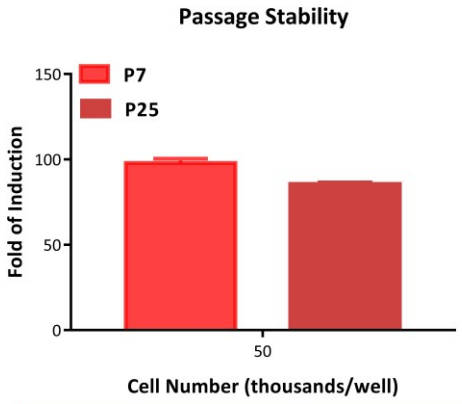


Fig3. Inhibition of human BTLA overexpressing on CHO cells induced reporter activity by anti-human BTLA antibody. This reporter cell was incubated with serial dilutions of antibodies in the presence of CHO/Human BTLA Stable Cell Line. The EC50 of anti-human BTLA neutralizing antibody is approximately 0.212 μg/mL.

• *Passage Stability*



	P7	P25
Max Fold	97.91	85.74

Fig4. Passage stability analysis by Signaling Bioassay. The continuously growing Human HVEM (Luc) HEK293 Reporter Cell was incubated with CHO/Human BTLA Stable Cell Line (Cat.No.SCCHO-ATP110). The human BTLA overexpressing on CHO cells stimulated response demonstrates passage stabilization (fold induction) across passage 7-25.

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• *License Disclosure*

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• *Related Products*

Products	Cat.No.
CHO/Human BTLA Stable Cell Line Development Service	SCCHO-ATP110
Human BTLA (Luc) Jurkat Reporter Cell Development Service	SCJUR-STF106