HEK293/Human TrkB Stable Cell Line Data Sheet

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Catalog No.	Size
CHEK-ATP082	2 × (1 vial contains ~5×10^6 cells)

• *Description*

The HEK293/Human TrkB Stable Cell Line was engineered to express the receptor full length human TrkB (Uniprot: Q16620-1). Surface expression of human TrkB was confirmed by flow cytometry.

• *Application*

- Useful for cell-based TrkB binding assay

• *Cell Line Profile*

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

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- ***Materials Required for Cell Culture***
  - DMEM Medium (BasalMedia, Cat. No. L120KJ)

    Note: If you are unable to obtain the specified DMEM medium (BasalMedia, Cat. No. L120KJ) in China, you may use an alternative DMEM medium (Gibco, Cat. No. 11965-092) or another suitable medium for culturing.
  - Fetal bovine serum (CellMax, Cat. No. SA211.02)
  - Puromycin (InvivoGen, Cat.No.ant-pr-5b)

    Note: For selection antibiotics, we highly recommend using the specified brand. The activity of antibiotics may vary between manufacturers, so if you choose to use a different brand, it is essential to validate whether the concentration recommended in the culture medium is suitable. Regardless of the brand used, we recommend maintaining a backup culture without selection antibiotics to avoid potential cell loss due to inappropriate antibiotic concentration.
  - 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%P/S
  - Culture Medium: DMEM + 10% FBS, Puromycin (2 μg/mL), 1%P/S
  - Freeze Medium: 90% FBS, 10% (V/V) DMSO
  - T-75 Culture flask (Corning, Cat. No. 430641)
  - Cryogenic storage vials (SARSTEDT, Cat. No. 72.379.007)
  - Thermostat water bath
  - Centrifuge (Cence, Model: L550)
  - Cell counter (MONWEI, Model: SmartCell200A Plus)
  - CO_2 Incubator (Thermo, Model: 3111)
  - Biological Safety Cabinet (Thermo, Model: 1389)

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• *Recovery*

1. Thaw the vial by gently agitating it in a 37°C water bath. To minimize the risk of contamination, ensure the cap remains out of the water. Thawing should be completed quickly, typically within 3-5 minutes.
2. After thawing, promptly remove the vial from the water bath and decontaminate it by spraying with 70% ethanol. From this point onward, all operations must be performed under strict aseptic conditions.
3. Transfer the contents of the vial to a centrifuge tube containing 4.0 mL of complete growth medium. Centrifuge at approximately 1000 rpm for 5 minutes.
4. Resuspend the cell pellet with 5 mL complete growth medium and transfer the cell suspension into a 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. Cell viability may be low after thawing, and full recovery may take up to a week. Monitor the cells daily until the culture reaches 80-90% confluency. At this point, remove and discard the spent medium. Avoid allowing the cells to become over-confluent to ensure optimal cell health.
2. Wash the cells once with sterile PBS. Avoid adding PBS directly onto the cell surface.
3. Add 2 mL of 0.25% Trypsin-EDTA to the T-75 flask. Place the flask at 37°C for 2-3 minutes, until 90% of the cells have detached. Monitor under a microscope to avoid over-trypsinization.
4. Add 6.0 to 8.0 mL of culture medium using a pipette and gently rinse the cells from the surface of the T-75 flask. Gently pipette up and down several times to achieve a single cell suspension without cell clumps.
5. Transfer appropriate aliquots of the cell suspension to a new T-75 flask. A subcultivation ratio of 1:4 to 1:8 is recommended. Adjust the ratio based on your specific culture system.
6. Incubate at 37℃ with 5% CO_2 incubator.
7. When the cell culture reaches 80-90% confluency, proceed to the next subculture. Avoid over-confluency, as this may negatively impact cell performance in subsequent passages.

Note: After recovery, maintain the cells for 1-2 passages in the complete growth medium not containing the selection marker, if the cells are in good condition, transition to the culture medium containing the selection marker during subculturing.

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• *Cryopreservation*

1. When the cell culture reaches 80-90% confluency, remove and discard the spent medium.
2. Wash the cells once with sterile PBS. Avoid adding PBS directly onto the cell surface.
3. Add 2 mL of 0.25% Trypsin-EDTA to the T-75 flask. Place the flask at 37°C for 2-3 minutes, until 90% of the cells have detached. Monitor under a microscope to avoid over-trypsinization.
4. Add 6.0 to 8.0 mL of complete growth medium using a pipette and gently rinse the cells from the surface of the T-75 flask. Gently pipette up and down several times to achieve a single cell suspension without cell clumps. Count the viable cells.
5. Transfer the cell suspension to a centrifuge tube. Centrifuge at 1000 rpm for 5 min at room temperature to pellet the cells.
6. After centrifugation, discard the supernatant. Resuspend the cells in ice cold freezing medium to a concentration of 5×10^6 to 1×10^7 cells/mL.
7. Aliquot the cell suspension into cryogenic storage vials. Place the vials in a programmable cooler or an insulated box placed in a –80°C freezer overnight, then transfer to liquid nitrogen storage for long-term storage.

Note: It is recommended to establish a cell bank at the earliest possible passage for long-term use.

• *Storage*

Cells must be received in a frozen state on dry ice and should be transferred to liquid nitrogen or a –80°C freezer immediately upon receipt. If stored in a –80°C freezer, it is recommended to limit the storage period to no more than two weeks. For long-term preservation, transfer the cells to liquid nitrogen is highly recommended.

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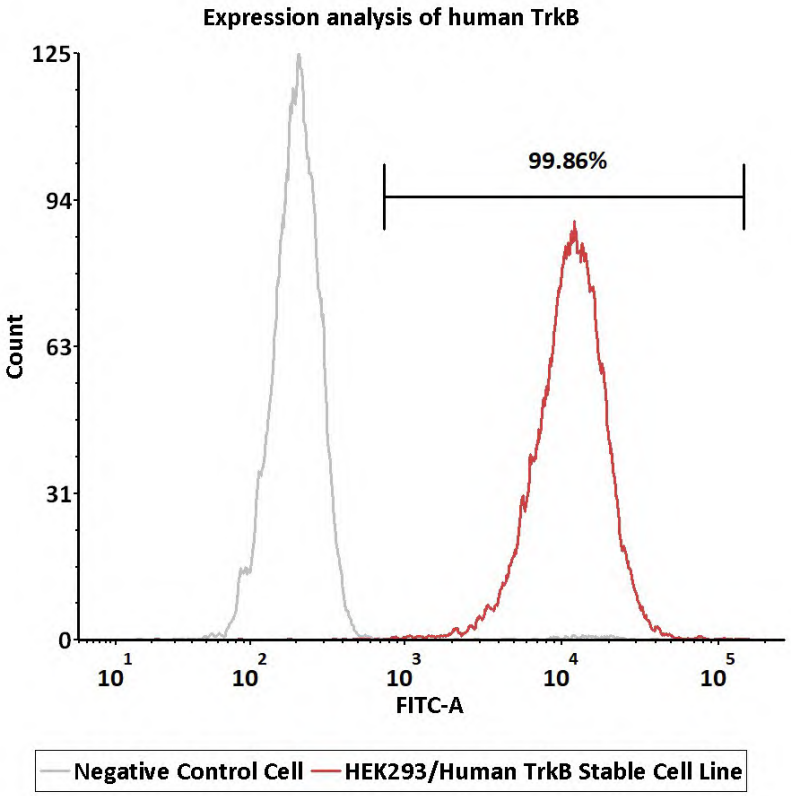
• ***Receptor Assay***



Catalog No.	Stable Cell Line	MFI for TrkB (FITC)
NA	Negative Control Cell	195.93
CHEK-ATP082	HEK293/ Human TrkB Stable Cell Line	11339.91

Fig1. Expression analysis of human TrkB on HEK293/Human TrkB Stable Cell Line by FACS. Cell surface staining was performed on HEK293/Human TrkB Stable Cell Line or negative control cell using anti-human TrkB antibody followed by staining with FITC anti-mouse IgG antibody.

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• *Related Products*

Products	Cat.No.
Human TrkA (Luc) HEK293 Reporter Cell	CHEK-ATF093
HEK293/Human APP (GFP) Stable Cell Line	CHEK-ATP081
HEK293/Human Alpha-synuclein (GFP) Stable Cell Line	CHEK-ATP085
HEK293/Human Tau-K18 (GFP) Stable Cell Line	CHEK-ATP087
Human 5-HT1A (Luc) HEK293 Reporter Cell	CHEK-ATF131
HEK293/Human SORT1 Stable Cell Line	CHEK-ATP155
HEK293/Human RAGE Stable Cell Line	CHEK-ATP156
HEK293/Human NGFR Stable Cell Line	CHEK-ATP157
HEK293/Human LDL R Stable Cell Line	CHEK-ATP158
HEK293/Human LILRB3 Stable Cell Line	CHEK-ATP159
Human CGRPR/RAMP1(Luc) HEK293 Reporter Cell	CHEK-ATF168
HEK293/Human TrkC Stable Cell Line	CHEK-ATP189
HEK293/Human TrkA Stable Cell Line	CHEK-ATP192