

HEK293/Human B7-H4 Stable Cell Line

| Catalog No. | Size |
|--------------|--|
| CHEK-ATP126- | $2 \times (1 \text{ vial contains } \sim 5 \times 10^{6} \text{ cells})$ |

• Description

The HEK293/Human B7-H4 Stable Cell Line was engineered to express the receptor full length human B7-H4 (Gene ID: 79679), used to mimic cancer target cells. Surface expression of human B7-H4 was confirmed by flow cytometry.

• Application

• Useful for cell-based B7-H4 binding assay

• Cell Line Profile

| Cell line | HEK293/Human B7-H4 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 | | |



• 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)
- 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, 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.

Note: After recovery for 1-2 generations with the complete growth medium not containing the selection marker, if the cell state is well, changing to the culture medium containing the selection marker.



• Cryopreservation

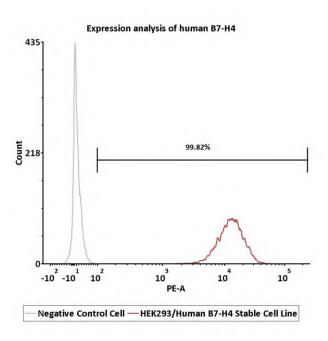
- 1. Remove and discard spent medium.
- 2. Detach cells from the cell culture flasks with 0.25% trypsin.
- 3. Centrifuge at 1000 rpm for 5 min at RT to pellet cells.
- 4. Resuspend the cell pellets with complete growth medium and count viable cells.
- 5. Centrifuge at 1000 rpm for 5 min at RT and resuspend 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



• Receptor Assay



| Catalog No. | Stable Cell Line | MFI for B7-H4 (PE) |
|-------------|-------------------------------------|--------------------|
| NA | Negative Control Cell | 4.59 |
| CHEK-ATP126 | HEK293/Human B7-H4 Stable Cell Line | 12332.57 |

Fig1. Expression analysis of human B7-H4 on HEK293/Human B7-H4 Stable Cell Line by FACS. Cell surface staining was performed on HEK293/Human B7-H4 Stable Cell Line or negative control cell using PE-labeled anti-human B7-H4 antibody.



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• Related Products

| <u>Products</u> | <u>Cat.No.</u> |
|---|----------------|
| HEK293/Human ROR1 Stable Cell Line | CHEK-ATP084 |
| HEK293/Human Mesothelin Stable Cell Line | CHEK-ATP119 |
| HEK293/Human Glypican-3 (GPC3) Stable Cell Line | CHEK-ATP092 |
| HEK293/Human FOLR1 Stable Cell Line | CHEK-ATP091 |
| HEK293/Human DLL3 Stable Cell Line | CHEK-ATP090 |
| HEK293/Human TL1A Stable Cell Line | CHEK-ATP142 |
| HEK293/Human NAPI-IIb Stable Cell Line | CHEK-ATP116 |
| HEK293/Human Cadherin-6 Stable Cell Line | CHEK-ATP127 |
| HEK293/Human ENPP3 Stable Cell Line | CHEK-ATP122 |