

## HEK293/Human GPR75 Stable Cell Line

Catalog No.	Size
CHEK-ATP174	$2 \times (1 \text{ vial contains } \sim 5 \times 10^6 \text{ cells})$

## • Description

The HEK293/Human GPR75 Stable Cell Line was engineered to express the receptor full length human GPR75 (Gene ID: 10936), used to mimic cancer target cells. Surface expression of human GPR75 was confirmed by flow cytometry.

## • Application

Useful for cell-based GPR75 binding assay

### • Cell Line Profile

Cell line	HEK293/Human GPR75 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 <sub>2</sub>	
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
- Culture Medium: DMEM + 10% FBS, Puromycin (2 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.

#### • 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

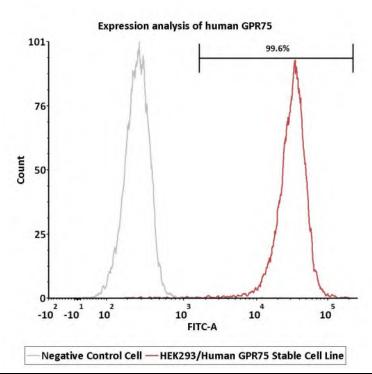
- 1. Remove and discard spent medium.
- 2. Detach cells from the cell culture flasks with 0.25% trypsin.
- 3. Centrifuge at 1000 rpm for 5 min at RT to pellet cells.
- 4. Resuspend the cell pellets with complete growth medium and count viable cells.
- 5. Centrifuge at 1000 rpm for 5 min at RT and resuspend cells in freezing medium to a concentration of  $5\times10^6$  to  $1\times10^7$  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 GPR75 (FITC)
NA	Negative Control Cell	258.92
CHEK-ATP174	HEK293/Human GPR75 Stable Cell Line	31511.14

**Fig1.** Expression analysis of human GPR75 on HEK293/Human GPR75 Stable Cell Line by FACS. Cell surface staining was performed on HEK293/Human GPR75 Stable Cell Line or negative control cell using anti-human GPR75 antibody followed by staining with FITC anti-mouse IgG antibody.



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#### • Related Products

<u>Products</u>	Cat.No.
HEK293/Human CEACAM5 Stable Cell Line	CHEK-ATP083
HEK293/Human DLL3 Stable Cell Line	CHEK-ATP090
HEK293/Human ROR1 Stable Cell Line	CHEK-ATP084
HEK293/Human FOLR1 Stable Cell Line	CHEK-ATP091
HEK293/Human Glypican-3 (GPC3) Stable Cell Line	CHEK-ATP092
CHO/Human Mesothelin Stable Cell Line Development Service	SCCHO-ATP120
CHO/Human Glypican-3 (GPC3) Stable Line Development Service	SCCHO-ATP112
CHO/Human STEAP1 Stable Cell Line Development Service	SCCHO-ATP121
CHO/Human uPAR Stable Cell Line	SCCHO-ATP152