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HEK293/Human GIPR Stable Cell Line (Low Expression)

Catalog No.	Size
CHEK-ATP208	$2 \times (1 \text{ vial contains} \sim 5 \times 10^{6} \text{ cells})$

• Description

The HEK293/Human GIPR Stable Cell Line was engineered to express the receptor full length human GIPR (Uniprot: P48546-1), with different levels of GIPR expression (High, Medium, Low). Surface expression of human GIPR was confirmed by flow cytometry.

• Application

- Useful for cell-based GIPR binding assay
- Screen for human GIPR agonists based on cAMP accumulation assay

• Cell Line Profile

Cell line	HEK293/Human GIPR Stable Cell Line (Low Expression)	
Host Cell	HEK293	
Property	Adherent	
Complete Growth Medium	DMEM + 10% FBS	
Selection Marker	Hygromycin B (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 (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)
- Hygromycin B (Invitrogen, Cat. No. 10687010)
- 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, Hygromycin B (20 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)
- CO2 Incubator (Thermo, Model: 3111)
- Biological Safety Cabinet (Thermo, Model: 1389)



• 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°C with 5% CO₂ 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°C with 5% CO₂ 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.



• Cryopreservation

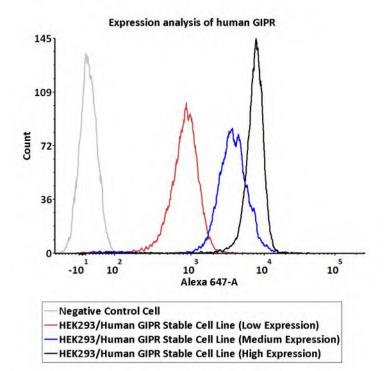
- 1. When the cell culture reaches 80-90% confluency, remove and discard the spent medium.
- 2. Wash the cells once with sterile PBS. Avoid adding PBS directly onto the cell surface.
- 3. Add 2 mL of 0.25% Trypsin-EDTA to the T-75 flask. Place the flask at 37°C for 2-3 minutes, until 90% of the cells 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.
- 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.



• Receptor Assay

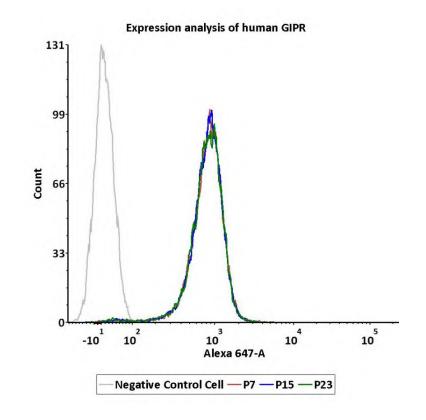


Catalog No.	Stable Cell Line	MFI for GIPR (Alexa 647)
CHEK-ATP208	HEK293/Human GIPR Stable Cell Line (Low Expression)	816.14
CHEK-ATP207	HEK293/Human GIPR Stable Cell Line (Medium Expression)	3519.54
CHEK-ATP206	HEK293/Human GIPR Stable Cell Line (High Expression)	7286.12

Fig1. Expression analysis of human GIPR on HEK293/Human GIPR Stable Cell Line by FACS. Cell surface staining using Alexa 647-labeled anti-human GIPR antibody was performed on HEK293/Human GIPR Stable Cell Line (Low Expression); HEK293/Human GIPR Stable Cell Line (Low Expression); HEK293/Human GIPR Stable Cell Line (Medium Expression); HEK293/Human GIPR Stable Cell Line (High Expression).



• Passage Stability



Passage	MFI for GIPR (Alexa 647)
P7	815.25
P15	812.61
P23	803.78

Fig2. Passage stability analysis of receptors expression by FACS. Flow cytometry surface staining of human GIPR on HEK293/Human GIPR Stable Cell Line (Low Expression) demonstrates consistent mean fluorescent intensity across passage 7-23.



Products_	<u>Cat.No.</u>
HEK293/Human GIPR Stable Cell Line (High Expression)	CHEK-ATP206
HEK293/Human GIPR Stable Cell Line (Medium Expression)	CHEK-ATP207
HEK293/Human GLP-1R Stable Cell Line (High Expression)	CHEK-ATP160
HEK293/Human GLP-1R Stable Cell Line (Medium Expression)	CHEK-ATP161
HEK293/Human GLP-1R Stable Cell Line (Low Expression)	CHEK-ATP162
HEK293/Human ASGR1 Stable Cell Line	CHEK-ATP080
Human GLP-1R (Luc) HEK293 Reporter Cell	CHEK-ATF096
Human GCGR (Luc) HEK293 Reporter Cell	CHEK-ATF103
Human GIPR (Luc) HEK293 Reporter Cell	CHEK-ATF104
Human FGF-21 (Luc) HEK293 Reporter Cell	CHEK-ATF163
Human Activin RII (Luc) HEK293 Reporter Cell	CHEK-ATF164
HEK293/Human ASGR1&ASGR2 Stable Cell Line	CHEK-ATP172
HEK293/Human GPR75 Stable Cell Line	CHEK-ATP174
Human THRB (Luc) HEK293 Reporter Cell	CHEK-ATF181
Human THRA (Luc) HEK293 Reporter Cell	CHEK-ATF180
HEK293/Human GLP-1R&GIPR Stable Cell Line	CHEK-ATP205