

# NIH-3T3/Human IGF-1 R Stable Cell Line Development Service

Catalog No.	Size	
CNIH-ATP102	$2 \times (1 \text{ vial contains} \sim 5 \times 10^{6} \text{ cells})$	

## • Description

NIH-3T3/Human IGF-1 R Stable Cell Line was engineered to express the receptor full length human IGF-1 R (Gene ID: 3480). Surface expression of Human IGF-1 R was confirmed by flow cytometry.

# • Application

• Useful for cell-based IGF-1 R binding assay

# • Cell Line Profile

Cell line	NIH-3T3/Human IGF-1 R Stable Cell Line		
Host Cell	NIH-3T3		
Property	Adherent		
Complete Growth Medium	DMEM + 10% NBCS		
Selection Marker	Hygromycin (20 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)
- Newborn calf serum (Gibco, Cat.No.16010-159)
- Hygromycin B (Invitrogen, Cat.No.10687010)
- Complete Growth Medium: DMEM + 10% NBCS
- Culture Medium: DMEM + 10% NBCS, 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
- MONWEI<sup>TM</sup>SmartCell200 (Shanghai Monwei Biomedical Technology Co. ,Ltd)
- 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. Discard the supernatant and 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  $37^{\circ}$ C with 5% CO<sub>2</sub> incubator.



## • Subculture

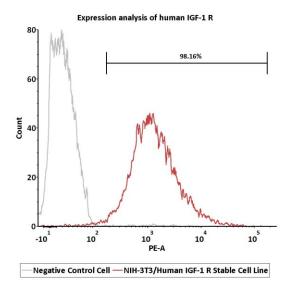
- 1. Remove and discard culture medium.
- 2. Wash the cells once with sterile PBS.
- Add 2 mL of 0.25% trypsin to cell culture flask. Place the flask at 37°C for 2-5 minutes, until 90% of the cells have detached
- 4. Add 6.0 to 8.0 mL of culture medium and aspirate cells by gently pipetting.
- 5. Add 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 1:4 to 1:8 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  $5 \times 10^6$  to  $1 \times 10^7$  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 IGF-1 R (PE)
NA	Negative Control Cell	17.93
CNIH-ATP102	NIH-3T3/Human IGF-1 R Stable Cell Line Development Service	1178.26

Fig1. Expression analysis of human IGF-1 R on NIH-3T3/Human IGF-1 R Stable Cell Line by FACS. Cell surface staining was performed on NIH-3T3/Human IGF-1 R Stable Cell Line or negative control cell using PE-labeled anti-human IGF-1 R antibody.

## • License Disclosure

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