

# Product Sheet

## EGFR C797S Homozygous RKO Cell Line

Catalog number: GM-C42673

Version 3.3.1.250822

EGFR is a member of the receptor tyrosine kinase (TK) family. It is widely distributed on the surface of cells such as mammalian fibroblasts and epithelial cells. The EGFR signaling pathway plays a crucial role in physiological processes like cell proliferation and differentiation. When EGFR mutates, it causes the EGFR signaling pathway to remain persistently activated without ligand binding, leading to abnormal cell proliferation.

EGFR C797S Homozygous RKO Cell Line is a clonal stable RKO cell line constructed using non-viral transfection. Using gene editing technology to introduce the C797S point mutation into the endogenous EGFR gene in cell lines.

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## Specifications

<b>Quantity</b>	5E6 Cells per vial, 1 mL
<b>Product Format</b>	1 vial of frozen cells
<b>Shipping</b>	Shipped on dry ice
<b>Storage Conditions</b>	Liquid nitrogen immediately upon receipt

<b>Recovery Medium</b>	DMEM+10% FBS+1% P.S
<b>Growth medium</b>	DMEM+10% FBS+1% P.S
<b>Note</b>	Cells contain Blasticidin resistance genes.
<b>Freezing Medium</b>	90% FBS+10% DMSO
<b>Growth properties</b>	Adherent
<b>Growth Conditions</b>	37°C, 5% CO <sub>2</sub>

<b>Mycoplasma Testing</b>	The cell line has been screened to confirm the absence of Mycoplasma species.
<b>Safety considerations</b>	Biosafety Level 2
<b>Note</b>	It is recommended to expand the cell culture and store a minimum of 10 vials at an early passage for potential future use.

## Materials

<b>Reagent</b>	<b>Manufacturer/Catalogue No.</b>
DMEM	Gibco/C11995500BT
Fetal Bovine Serum	ExCell/FSP500
Pen/Strep	Thermo/15140-122

## Figures

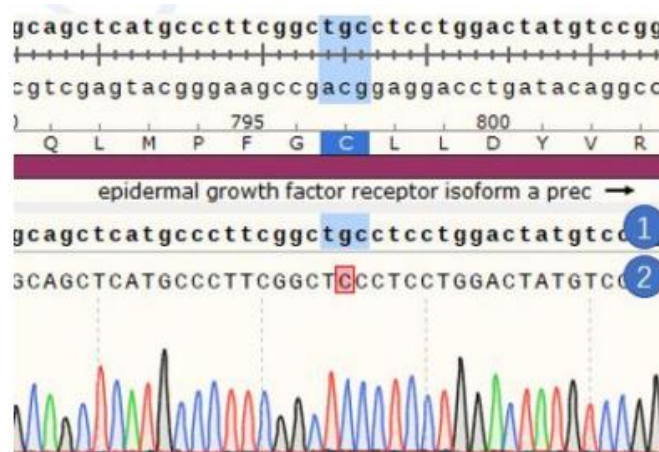


Figure 1 | The EGFR mutation analysis by Sanger sequencing. 1 represents the wild-type sequence, and 2 represents the sequence after knock-in mutation.

## Cell Recovery

Recovery Medium: DMEM+10% FBS+1% P.S

To insure the highest level of viability, thaw the vial and initiate the culture as soon as possible upon receipt. If upon arrival, continued storage of the frozen culture is necessary, it should be stored in liquid nitrogen vapor phase and not at -70°C. Storage at -70°C will result in loss of viability.

- Thaw the vial by gentle agitation in a 37°C water bath. To reduce the possibility of contamination, keep the O-ring and cap out of the water. Thawing should be rapid (approximately 2 - 3 minutes).
- Remove the vial from the water bath as soon as the contents are thawed, and decontaminate by dipping in or spraying with 70% ethanol. All of the operations from this point on should be carried out under strict aseptic conditions.
- Transfer the vial contents to a centrifuge tube containing 5.0 mL complete culture medium and spin at approximately 176 x g for 5 minutes. Discard supernatant.
- Resuspend cell pellet with the recommended recovery medium. And dispense into appropriate culture dishes.
- Incubate the culture at 37°C in a suitable incubator. A 5% CO<sub>2</sub> in air atmosphere is recommended if using the medium described on this product sheet.

## Cell Freezing

Freezing Medium: 90% FBS+10% DMSO

- Centrifuge at 176 x g for 3 minutes to collect cells.
- Resuspend the cells in pre-cooled freezing medium and adjust the cell density to 5E6 cells/mL.
- Aliquot 1 mL into each vial.
- Place the vial in a controlled-rate freezing container and store at -80°C for at least 1 day, then transfer to liquid nitrogen as soon as possible.

## Cell passage

Growth medium: DMEM+10% FBS+1% P.S

For the first 1 to 2 passages post-resuscitation, use the recovery medium. Once the cells have stabilized, switch to a growth medium.

- a) Remove and discard culture medium.
- b) Briefly rinse the cell layer with PBS to remove all traces of serum that contains trypsin inhibitor.
- c) Add 1.0 mL of 0.25% (w/v) Trypsin-EDTA solution to dish and observe cells under an inverted microscope until cell layer is dispersed (usually within 30 to 60 seconds at 37°C).
- d) Note: To avoid clumping do not agitate the cells by hitting or shaking the flask while waiting for the cells to detach. Cells that are difficult to detach may be placed at 37°C to facilitate dispersal.
- e) Add 2.0 mL of growth medium to mix well and aspirate cells by gently pipetting.
- f) After centrifugation, resuspend the pellet and add appropriate aliquots of the cell suspension to new culture vessels.
- g) Incubate cultures at 37°C.

**Subcultivation Ratio: A subcultivation ratio of 1:3 - 1:4 is recommended**

**Medium Renewal: Every 2 to 3 days**

## Notes

- a) It is normal to observe a higher number of dead cells immediately after thawing. The condition will improve significantly after adjustment. Once the cells stabilize, the number of dead cells will decrease after subculturing, and the cell growth rate will become stable.

## License Agreement:

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