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Product Sheet

BRCA2 KO HCT116 Cell Line

Catalog number: GM-C33656

Version 3.3.1.250415

Description	BRCA2 KO HCT116 Cell Line is a clonal stable cell line derived from HCT116 cells with a knockout of BRCA2.	
Quantity	5E6 Cells per vial,1 mL	
Product Format	1 vial of frozen cells	
Shipping	Shipped on dry ice	
Storage Conditions	Liquid nitrogen immediately upon receipt	
Target	Human BRCA2	
Gene ID/Uniprot ID		
Host Cell	HCT116	
Recovery Medium	McCoy's 5A+10% FBS+1% P.S	
Growth medium	McCoy's 5A+10% FBS+1% P.S	
Note	The cells are very sensitive to antibiotics and should be cultured using the cell growth medium provided by Genomeditech. These cells are constructed based on BRCA2 knockout parent cells, which contain Blasticidin resistance genes.	
Freezing Medium	90% FBS+10% DMSO	
Growth properties	Adherent	
Growth Conditions	37°C, 5% CO ₂	
Mycoplasma Testing	The cell line has been screened to confirm the absence of Mycoplasma species.	
Safety considerations	Biosafety Level 2	
Note	It is recommended to expand the cell culture and store a minimum of 10 vials at an early passage for potential future use.	

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Materials

Reagent	Manufacturer/Catalogue No.
McCoy's 5A	VivaCell/C3020-0500
Fetal Bovine Serum	Cegrogen biotech/A0500-3010
Pen/Strep	Thermo/15140-122

Figures

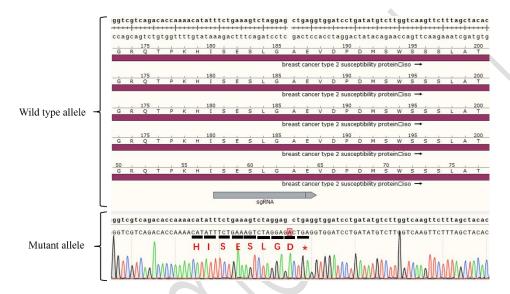


Figure 1 | The Sanger sequencing of the BRCA2 KO HCT116 Cell Line showed successful knockout of BRCA2.

Cell Recovery

Recovery Medium: McCoy's 5A+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.

- a) 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).
- b) 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.
- c) 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.
- d) Resuspend cell pellet with the recommended recovery medium. And dispense into appropriate culture dishes.
- e) Incubate the culture at 37°C in a suitable incubator. A 5% CO₂ in air atmosphere is recommended if using the medium described on this product sheet.

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Cell Freezing

Freezing Medium: 90% FBS+10% DMSO

- a) Centrifuge at 176 x g for 3 minutes to collect cells.
- b) Resuspend the cells in pre-cooled freezing medium and adjust the cell density to 5E6 cells/mL.
- c) Aliquot 1 mL into each vial.
- d) 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: McCoy's 5A+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 1 to 2 minutes at 37°C).
- 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 1 to 2 days

Notes

a) After the stabilization of the cell condition, there will be fewer dead cells post-passage, the cell growth rate will tend to stabilize, cell morphology will become uniform, and the cells will appear robust.

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