

OncoSpot™ KRAS G13C/G13C HCT116 Cancer Mutation Cell Line

Catalog number: SL710

Product: KRAS G13C HCT116 cancer mutation cell line (homozygote)

Description: This product is a HCT116 cell line genetically modified using CRISPR to have

both alleles with a G->T point mutation at the KRAS gene position 37 in exon 2

and a puromycin marker in intron 1 of KRAS for selection.

Genotype: KRAS (G13C, puro/ G13C, puro)

Genomic Mutation: gttggagctggtGgc -> gttggagctggtTgc

Quantity: 1 vial of 2 x 10⁶ cells; frozen

Shipping conditions: Dry ice

Storage conditions: Liquid nitrogen vapor phase. Remove the item from the dry ice packaging

and check all items for damage and leakage. Place immediately into storage at

or below -140 °C, preferably into the liquid nitrogen vapor phase, until use.

Source of parental line:

HCT116

Organism: Homo sapiens, human

Tissue: colon

Cell Type: epithelial

Quality control: >95% viability before freezing. All cells were tested and found to be free

of mycoplasma, bacteria, viruses, and other toxins.

Safety instructions: To ensure safety, protective gloves, clothing, and a face mask should be worn

when handling frozen vials. Some leakage may occur into the vial during storage. The liquid nitrogen will be converted to gas upon thawing. Due to the nature of nitrogen gas, pressure may build within the vial and possibly result in

the vial exploding or losing its cap. This may cause flying debris.

Thawing procedure: The vial of cells should be thawed in a 37 °C water bath with gentle agitation.

For optimal performance, the vial should be thawed in under two minutes. Ensure that the cap of the vial did not loosen upon thawing, and re-tighten as



needed. Spray the vial with 70% EtOH and wipe off. Repeat. Using aseptic technique, add the contents of the vial to 9 ml of complete growth medium (without selection). Centrifuge for 5 min. at 250 xg. Aspirate the medium, being careful not to disturb the pellet. Resuspend in 10 mL of complete growth medium, and place into a culture vessel of your choice. Only add selection to the medium after 24 hours in culture.

Culture conditions: Complete Growth Medium

The base medium for this cell line is RPMI1640. For optimal growth and maintenance of selection, add the following components to the base medium: fetal bovine serum to a final concentration of 10%.

Selection

Puromycin to a final concentration of 0.6 μg/mL.

Culture temperature

37 °C with 5% CO_{2.}

Subculture

Replace culture medium with selection-free medium and incubate for up to 6 hours. Rinse the cells and split at 1×10^5 viable cells/mL to 1×10^6 cells/mL.

Cryopreservation

Freeze slowly in complete growth medium supplemented with 5% (v/v) DMSO.

Mycoplasma

Negative (MycoAlert Mycoplasma Detection Kit from Lonza).



Product Quality Control

FISH of KRAS probes on HCT116 cell lines

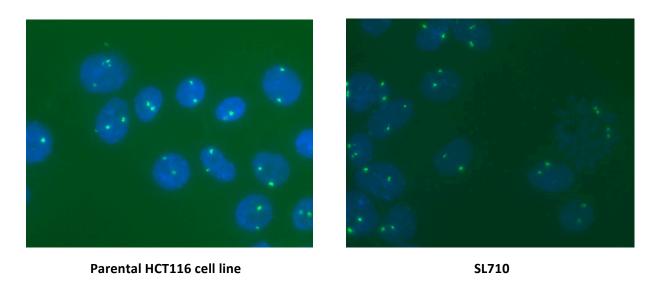
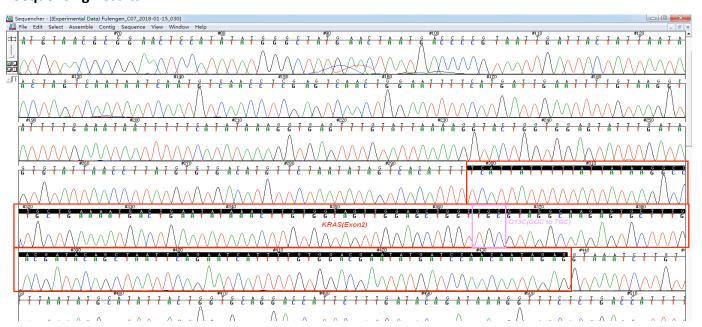


Figure 1. FISH of the human HCT116 cell lines. GCI VividFISH FISH probes CEP12 were hybridized in HCT116 cell line and SL710 mutant cell line. The two green signals are from chromosome 12 centromeres. The nuclei are counter stained with DAPI (blue).

Sequencing Results





Citation of product: If use of this item results in a publication, please use this information:

OncoSpot™ KRAS G13C/G13C HCT116 cancer mutation cell line (SL710,

GeneCopoeia, Inc., Rockville, MD).

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