Product sheet

B-LCL-HROC59 Cells | 302073

General information

Description	B-LCL-HROC59 is an Epstein-Barr virus (EBV)-immortalized human B lymphoblastoid cell line generated from tumor-infiltrating B cells (TiBc) isolated from a primary colorectal carcinoma designated HROC59. The parental tumor was resected from an adult male patient with right-sided sporadic colorectal carcinoma and advanced-stage disease. Fresh tumor tissue was mechanically dissociated to obtain single-cell suspensions, and B cells were selectively immortalized in vitro using EBV-containing supernatant derived from the B95/8 marmoset cell line in the presence of cyclosporin A to suppress T- and NK-cell expansion. Long-term culture resulted in stable outgrowth of a monoclonal B-cell population, as demonstrated by immunoglobulin gene rearrangement analysis. B-LCL-HROC59 secretes immunoglobulin G (IgG) as its exclusive isotype, with stable production over prolonged culture. In cell-based binding assays, IgG derived from B-LCL-HROC59 demonstrated only minimal binding to tested allogeneic colorectal carcinoma cell lines compared with other TiBc-derived IgGs exhibiting stronger tumor cell reactivity. No evidence of spontaneous B-cell outgrowth was observed in the absence of exogenous EBV during culture establishment, indicating that immortalization occurred in vitro rather than reflecting latent EBV-driven transformation in vivo. As a monoclonal, antigen-experienced tumor-infiltrating B-cell line, B-LCL-HROC59 provides a defined model for studying humoral immune responses within the colorectal cancer microenvironment and for investigating the specificity and functional properties of tumor-associated antibodies.
Organism	Human
Tissue	Peripheral blood
Disease	Carcinoma
Synonyms	Bc HROC59, TiBcHROC59

Characteristics

Age	76 years
Gender	Male
Ethnicity	Caucasian
Morphology	Round cells
Cell type	B lymphoblast
Growth properties	Suspension

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Regulatory Data

Citation	B-LCL-HROC59 (Cytion catalog number 302073)
Biosafety level	2
NCBI_TaxID	9606
CellosaurusAccession	CVCL_A7US

Biomolecular Data

Surface antigens	CD19
Viruses	Transformant: EBV

Handling

Culture Medium	RPMI 1640, w: 2.0 mM stable Glutamine, w: 2.0 g/L NaHCO3 (Cytion article number 820700a)
Supplements	Supplement the medium with 10% heat-inactivated FBS
Subculturing	Gently homogenize the cell suspension in the flask by pipetting up and down, then take a representative sample to determine the cell density per ml. Dilute the suspension to achieve a cell concentration of 1 x 10^5 cells/ml with fresh culture medium, and aliquot the adjusted suspension into new flasks for further cultivation.
Freeze medium	As a cryopreservation medium, we use complete growth medium (including FBS) + 10% DMSO for adequate post-thaw viability, or CM-1 (Cytion catalog number 800100), which includes optimized osmoprotectants and metabolic stabilizers to enhance recovery and reduce cryo-induced stress.

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Thawing and Culturing Cells	1. Confirm that the vial remains deeply frozen upon delivery, as cells are shipped on dry ice to maintain optimal temperatures during transit. 2. Upon receipt, either store the cryovial immediately at temperatures below -150°C to ensure the preservation of cellular integrity, or proceed to step 3 if immediate culturing is required. 3. For immediate culturing, swiftly thaw the vial by immersing it in a 37°C water bath with clean water and an antimicrobial agent, agitating gently for 40-60 seconds until a small ice clump remains. 4. Perform all subsequent steps under sterile conditions in a flow hood, disinfecting the cryovial with 70% ethanol before opening. 5. Carefully open the disinfected vial and transfer the cell suspension into a 15 ml centrifuge tube containing 8 ml of room-temperature culture medium, mixing gently. 6. Centrifuge the mixture at 300 x g for 3 minutes to separate the cells and carefully discard the supernatant containing residual freezing medium. 7. Gently resuspend the cell pellet in 10 ml of fresh culture medium. For adherent cells, divide the suspension between two T25 culture flasks; for suspension cultures, transfer all the medium into one T25 flask to promote effective cell interaction and growth. 8. Adhere to established subculture protocols for continued growth and maintenance of the cell line, ensuring reliable experimental outcomes.
Incubation Atmosphere	37°C, 5% CO_2, humidified atmosphere.
Shipping Conditions	Cryopreserved cell lines are shipped on dry ice in validated, insulated packaging with sufficient refrigerant to maintain approximately −78 °C throughout transit. On receipt, inspect the container immediately and transfer vials without delay to appropriate storage.
Storage Conditions	For long-term preservation, place vials in vapor-phase liquid nitrogen at about −150 to −196 °C. Storage at −80 °C is acceptable only as a short interim step before transfer to liquid nitrogen.

Quality Control & Molecular Analysis

Sterility	Mycoplasma contamination is excluded using both PCR-based assays and luminescence-based mycoplasma detection methods. To ensure there is no bacterial, fungal, or yeast contamination, cell cultures are subjected to daily visual inspections.