

Wilms11 Cells | 300420

Informações gerais

Description

The Wilms11 cell line was derived from a primary Wilms tumor (nephroblastoma) in a pediatric patient. Unlike many other Wilms tumor cell lines, Wilms11 is characterized by the presence of wild-type WT1, meaning it does not harbor mutations in the WT1 gene, which is typically associated with Wilms tumors exhibiting more aggressive or stromal phenotypes. However, the Wilms11 tumor exhibited significant stromal differentiation, with large areas of rhabdomyomatous differentiation, indicative of mesenchymal elements within the tumor. The presence of wild-type WT1, coupled with the tumor’s stromal differentiation, provides a unique model for understanding Wilms tumor biology in cases where WT1 mutations are absent.

Genetic studies of Wilms11 have shown that this cell line carries a tumor-specific mutation in CTNNB1, the gene encoding β -Catenin, which plays a crucial role in the Wnt signaling pathway. In Wilms11, this mutation affects serine 45, a key phosphorylation site involved in β -Catenin degradation. The CTNNB1 mutation results in the stabilization of β -Catenin, leading to its accumulation and constitutive activation of the Wnt signaling pathway, a driver of cell proliferation and tumorigenesis. This makes Wilms11 an important model for studying the interplay between Wnt signaling and Wilms tumor development, particularly in cases where WT1 remains intact.

Proteomic analyses of Wilms11 have revealed activation of several receptor tyrosine kinases (RTKs), including PDGFR β and AXL, which are involved in driving tumor cell growth and survival. Downstream signaling pathways, such as the MAPK and PI3K/AKT pathways, are also activated in Wilms11 cells, contributing to their tumorigenic behavior. The ability of Wilms11 cells to undergo mesenchymal differentiation, particularly rhabdomyomatous differentiation, highlights their potential as a model for studying the mesenchymal components of Wilms tumor. Overall, Wilms11 serves as a valuable tool for investigating the molecular mechanisms that drive Wilms tumorigenesis in the absence of WT1 mutations but in the context of Wnt pathway activation.

Organism Human

Tissue Kidney

Disease Wilms tumor

Applications In vitro cell culture model. Biochemical studies

Características

Age 22 months

Gender Male

Ethnicity Caucasian

Morphology Spindle-shaped

Cell type Wilms cells

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Growth properties Adherent

Dados regulatórios

Citation Wilms11 (Cytion catalog number 300420)

Biosafety level 1

NCBI_TaxID 9606

CellosaurusAccession CVCL_A5SM

Dados biomoleculares

Mutational profile WT1 mutation status: homozygous WT1 c.901c>T, p.R301x. LOH: . CTNNB1 mutation status: wild type

Manuseio

Culture Medium MSCGM kit (from Lonza)

Dissociation Reagent Accutase

Subculturing Remove the old medium from the adherent cells and wash them with PBS that lacks calcium and magnesium. For T25 flasks, use 3-5 ml of PBS, and for T75 flasks, use 5-10 ml. Then, cover the cells completely with Accutase, using 1-2 ml for T25 flasks and 2.5 ml for T75 flasks. Let the cells incubate at room temperature for 8-10 minutes to detach them. After incubation, gently mix the cells with 10 ml of medium to resuspend them, then centrifuge at 300xg for 3 minutes. Discard the supernatant, resuspend the cells in fresh medium, and transfer them into new flasks that already contain fresh medium.

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 $300 \times g$ 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.

Controle de Qualidade e Análise Molecular

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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.