Product sheet

Wilms1 Cells | 300411

General information

Description	The patient presented with bilateral tumors, not responding to the SIOP-2001 pre-surgery chemotherapy (4 weeks of Act D and VCR), the tumors were removed by kidney sparing surgery. In the following year without cytotoxic treatment, the patient again developed bilateal tumors. Cell culture was established from the left tumor which showed fetal rhabdomyomatous histology as described by Royer-Pokora 2010.
Organism	Human
Tissue	Kidney
Applications	In vitro cell culture model. Biochemical studies
Synonyms	Wilms1-2l

Characteristics

Age	2 years
Gender	Female
Ethnicity	Caucasian
Morphology	Spindle-shaped
Cell type	Wilms cells
Growth properties	Adherent

Identifiers / Biosafety / Citation

Citation	Wilms1 (Cytion catalog number 300411)
Biosafety level	1
Depositor	B. Royer-Pokora

Expression / Mutation

Receptors expressed	Receptor tyrosine kinases EGFR, EphA7, PDGFRalpha, FGFR1, PDGFRbeta, AxL
Tumorigenic	Yes, in nude mice. Forms tumor with small cells consistent with Wilms' tumor (xenografts may not represent Wilm?s tumors completely, see E. Kunce Stroup 2017)
Viruses	HIV-1: negative, HBV: negative, HCV: negative
Mutational profile	WT1 mutation status: homozygous c. 149 C>A, p.S50x, LOH: 11p11-11pter, CTNNB1 mutation status: heterozygous TCT>TTT, p.S45F
Karyotype	46, normal

Handling

Culture Medium	MSCGM kit (from Lonza)
Passaging solution	Accutase
Doubling time	24 hours
Subculturing	Remove medium and rinse the adherent cells using PBS without calcium and magnesium (3-5 ml PBS for T25, 5-10ml for T75 cell culture flasks). Add Accutase (1-2ml per T25, 2.5ml per T75 cell culture flask), the cell sheet must be covered completely. Incubate at ambient temperature for 10 minutes. Carefully resuspend the cells with medium (10 ml), centrifuge for 3 min at 300 g, resuspend cells in fresh medium and dispense into new flasks which contain fresh medium.
Seeding density	1 x 10^4 cells/cm^2
Fluid renewal	1 to 2 times per week
Freezing recovery	Fast
Freeze medium	CM-1 (Cytion catalog number 800100) or CM-ACF (Cytion catalog number 806100)

Handling of cryopreserved cultures	Wilms1 cells are shipped in a deep-frozen state on dry ice. Upon receipt, confirm that the vial remains frozen. For storage, place the cryovial immediately at temperatures below -150 degrees. If you plan to culture the cells immediately, swiftly thaw the vial by shaking it in a 37 degrees water bath with clean water and an antimicrobial agent for 40-60 seconds. Remove the vial once a small ice clump persists, ensuring it remains cold. Proceed with all subsequent steps under aseptic conditions. In a sterile flow hood, disinfect the cryovial with 70% ethanol. Then, gently open the vial and transfer the cell suspension into a 15 ml centrifuge tube pre-filled with 8 ml of room temperature culture medium. Gently mix the cells. For cell separation, centrifuge at 300 x g for 3 minutes and dispose of the supernatant. Skipping centrifugation is optional, although any residual freezing medium should be removed after 24 hours. Resuspend the pellet gently in 10 ml of fresh culture medium and divide between two T25 culture flasks. Follow the subculture protocol for subsequent steps.
Handling of proliferating cultures	One or two cell culture flasks come filled with cell culture medium. Collect the entire medium in a 50 ml centrifuge tube. Spin down the collected medium at 300 x g for 3 minutes to collect the cells which may have detached during transit. If a cell pellet is visible, resuspend the cells in 5 ml of cell culture medium and transfer to a T25 cell culture flask. Carefully add 5 ml of cell culture medium to each T25 cell culture flask. Examine cell morphology and confluency using a microscope. Finally, incubate the flasks at 37 degrees Celsius for at least 24 hours.

Quality control / Genetic profile / HLA

Sterility	Mycoplasma contamination is rigorously 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.
STR profile	**Amelogenin**: x,x **CSF1PO**: 10,12 **D13S317**: 11,13 **D16S539**: 11,14 **D5S818**: 12,13,14 **D7S820**: 9,14 **TH01**: 09. Mrz **TPOX**: 8,9 **vWA**: 14,19 **D3S1358**: 14,17,18 **D21S11**: 30,31 **D18S51**: 15,18 **Penta E**: 5,14 **Penta D**: 13 **D8S1179**: 12,14 **FGA**: 22,25

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HLA alleles	**A***: 03:01:01, 24:02:01 **B***: 35:03:01, 38:01:01 **C***: 12:03:01 **DRB1***: 07:01:01, 14:54:01 **DQA1***: 01:04:01, 02:01:01 **DQB1***: 02:02:01, 05:03:01 **DPB1***: 02:01:02G, 04:02:01G **E**: 01:03:01, 01:03:02