L-WRN

300641

GENERAL INFORMATION

Description

The L-WRN cell line is a murine fibroblast cell line derived from the L cells, which are mouse fibroblasts originally isolated from connective tissue. L-WRN cells have been engineered to stably express Wnt3a, R-spondin 3, and Noggin. These factors are critical for the growth and maintenance of intestinal organoids and stem cell cultures. The overexpression of these proteins enhances the proliferation and differentiation of intestinal stem cells, making L-WRN cells a valuable tool for studying intestinal biology and disease modeling.
In addition to their application in organoid culture, L-WRN cells serve as a robust model for investigating Wnt signaling pathways. Wnt signaling is pivotal in regulating cell fate, proliferation, and migration during development and in adult tissues. By providing a consistent and controlled source of Wnt3a, R-spondin 3, and Noggin, L-WRN cells facilitate research into the molecular mechanisms underlying these processes. Researchers can use these cells to dissect the roles of these signaling molecules in various biological contexts, including cancer, tissue regeneration, and developmental biology.
Overall, the L-WRN cell line is a powerful tool in biomedical research due to its ability to support the growth of complex three-dimensional cultures and its utility in studying key signaling pathways. Its role in the advancement of intestinal stem cell research and its contributions to our understanding of Wnt signaling highlight its importance in the field of cellular and molecular biology.

Tissue	Connective tissue

Applications

3D cell culture

CLASSIFICATION

Organism	Mouse
Sex	Male
Growth properties	Adherent
GMO status	GMO-S1

GMO status details

GMO-S1: This murine NIH-3T3-derived cell line (L-WRN) contains expression constructs for Wnt3a, R-spondin-3, and Noggin, including SV40 DNA sequences and dual antibiotic markers (hph and Tn5-neo), enabling secretion of these signaling molecules. The inserts are stably present in NIH-3T3-based cells. This classification applies only within Germany and may differ elsewhere.

Biosafety level	1

CHARACTERISTICS

Morphology	Fibroblast

IDENTIFIERS

Citation

L-WRN (Cytion catalog number 300641)

NCBI TaxID	10090
Cellosaurus	CVCL_DA06

Cytion GmbH
Nikola-Tesla-Str. 3
69124 Heidelberg
Tel: +49 (0)6221 405780
info@cytion.com
cytion.com

Registered office Heidelberg
VAT ID: DE283292917
Local court Mannheim - HRB 714394
CEO / Geschäftsführer: Jonathan Steubing

Banking information
Sparkasse Heidelberg
IBAN: DE35 6725 0020 0009 2076 86
BIC: SOLADES1HDB

EXPRESSION AND SUSCEPTIBILITY

Protein expression

Wnt-3A, R-spondin, noggin

HANDLING AND CULTURE

Culture medium

DMEM, w: 4.5 g/L Glucose, w: 4 mM L-Glutamine, w: 3.7 g/L NaHCO3, w: 1.0 mM Sodium pyruvate (Cytion article number 820300a)

Incubation atmosphere	37°C, 5% CO_2, humidified atmosphere.

Supplements

Supplement the medium with 10% FBS

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.

Thawing and culturing

Confirm that the vial remains deeply frozen upon delivery, as cells are shipped on dry ice to maintain optimal temperatures during transit.
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.
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.
Perform all subsequent steps under sterile conditions in a flow hood, disinfecting the cryovial with 70% ethanol before opening.
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.
Centrifuge the mixture at 300 x g for 3 minutes to separate the cells and carefully discard the supernatant containing residual freezing medium.
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.
Adhere to established subculture protocols for continued growth and maintenance of the cell line, ensuring reliable experimental outcomes.

QUALITY CONTROL AND CRYOPRESERVATION

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.

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.

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.

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.

DONOR

Breed / subspecies	C3H/An
Donor age	100 days

LEGAL AND SAFETY

Safety precautions

When planning to store a cryovial in liquid nitrogen for future thawing, it is mandatory to adhere to stringent safety measures. Appropriate protective gloves and clothing are essential, and the use of a face mask or safety goggles is required during the transfer of frozen samples to or from the liquid nitrogen tank. This is to mitigate the risk of injury from potential cryovial explosions upon removal, which can result in the projection of sharp fragments.

Warranty

At Cytion, we stand behind the quality and viability of our products.
We guarantee that our cells will be viable at the start of culture for
60 days after shipment from our facilities.
We assure that all materials conform to the specifications described in the
Certificate of Analysis and/or Product Sheet.
For full details, please refer to our
Warranty Statement.

Material Transfer Agreement required	Yes

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