

## MML-1 Cells | 300288

## General information

## Description

The MML-1 cell line is a melanoma cell line derived from malignant melanoma. This cell line is used primarily to study melanoma biology, particularly the role of extracellular vesicles (EVs) in cell-to-cell communication and tumor progression. MML-1 cells release various subtypes of EVs, including exosomes, microvesicles, and apoptotic bodies, each carrying distinct RNA cargos, such as microRNAs (miRNAs) and other non-coding RNAs.

Studies using MML-1 cells have demonstrated that exosomes released from these cells contain specific miRNAs like miR-214-3p, miR-199a-3p, and miR-155-5p, which are closely associated with melanoma progression and metastasis. These miRNAs are enriched in exosomes compared to other EV types and have been linked to important melanoma-related pathways, such as the regulation of the MAPK signaling pathway and tumor microenvironment interactions. Interestingly, comparisons of miRNA profiles from MML-1-derived exosomes with clinical samples of melanoma show a significant overlap, indicating the clinical relevance of this cell model in understanding melanoma progression.

In addition to miRNAs, MML-1 cells also release other non-coding RNAs such as small nucleolar RNAs (snoRNAs) and mitochondrial-associated transfer RNAs (mt-RNAs), which are differentially distributed among the EV subtypes. These findings highlight the MML-1 cell line's utility in studying the molecular mechanisms of melanoma, particularly how tumor cells communicate via EVs and influence their microenvironment.

**Organism** Human

**Tissue** Skin

**Disease** Melanoma

**Synonyms** MML1

## Characteristics

**Age** Unspecified

**Gender** Unspecified

**Morphology** Epithelial-like

**Growth properties** Adherent

## Regulatory Data

**Citation** MML-1 (Cytion catalog number 300288)

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<b>Biosafety level</b>	1
<b>NCBI_TaxID</b>	9606
<b>CellosaurusAccession</b>	CVCL_6004

**Biomolecular Data**

**Protein expression** P53 positive

**Tumorigenic** Yes, in nude mice

**Reverse transcriptase** Negative

**Mutational profile** V600E type BRAF Mutation was determined by DNA based methods (sequencing, RT-PCR) and protein based methods (Western Blot).

**Handling**

**Culture Medium** RPMI 1640, w: 2.0 mM stable Glutamine, w: 2.0 g/L NaHCO<sub>3</sub> (Cytion article number 820700a)

**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.

**Seeding density**  $1 \times 10^4$  cells/cm<sup>2</sup>

**Fluid renewal** 2 to 3 times per week

**Post-Thaw Recovery** After thawing, plate the cells at  $5 \times 10^4$  cells/cm<sup>2</sup> and allow the cells to recover from the freezing process and to adhere for at least 24 hours.

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### 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.

### 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<sub>2</sub>, 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.