

**M14 Cells | 302163**

**Renseignements généraux**

**Description**

The M14 cell line is a human melanoma cell line derived from a metastatic skin lesion of an adult patient with melanoma. This cell line is widely used in cancer research, particularly in the study of melanoma biology, tumor progression, and the evaluation of potential therapeutic agents. M14 cells exhibit characteristics typical of malignant melanoma, including the ability to form tumors in immunocompromised mice, making them a valuable tool for in vivo studies in addition to in vitro experiments.

In terms of molecular features, M14 cells have been reported to carry mutations in genes that are frequently altered in melanoma, including the BRAF gene. Specifically, M14 cells harbor the BRAF V600E mutation, which leads to constitutive activation of the MAPK/ERK signaling pathway, promoting cell proliferation and survival. This makes M14 an important model for studying targeted therapies, such as BRAF inhibitors, that are designed to exploit this mutation. Additionally, M14 cells have been utilized in immunotherapy research due to their expression of various melanoma-associated antigens and susceptibility to immune system modulation.

Researchers using the M14 cell line should note that these cells are not suitable for therapeutic applications and are intended solely for research purposes, particularly those focusing on melanoma pathophysiology, drug screening, and the development of new therapeutic strategies. The M14 cell line remains a key resource for advancing our understanding of melanoma and exploring new avenues for treatment.

**Organism** Human

**Tissue** Skin

**Disease** Amelanotic melanoma

**Metastatic site** Right buttock, hypodermis

**Synonyms** M14-MEL, UCLA-SO-M14, UCLA SO M14, UCLA-SO-14, UCLASO-M14, Melanoma 14, M-14

**Caractéristiques**

**Age** 33

**Gender** Male

**Ethnicity** European

**Morphology** Fibroblast-like

**Growth properties** Adherent

**M14 Cells | 302163****Données réglementaires****Citation** M14 (Cytion catalog number 302163)**Biosafety level** 1**NCBI\_TaxID** 9606**CellosaurusAccession** CVCL\_1395**Données biomoléculaires****Manipulation****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% heat-inactivated 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.**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  $-150^{\circ}\text{C}$  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  $37^{\circ}\text{C}$  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

$37^{\circ}\text{C}$ , 5%  $\text{CO}_2$ , humidified atmosphere.

### Shipping Conditions

Cryopreserved cell lines are shipped on dry ice in validated, insulated packaging with sufficient refrigerant to maintain approximately  $-78^{\circ}\text{C}$  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  $-196^{\circ}\text{C}$ . Storage at  $-80^{\circ}\text{C}$  is acceptable only as a short interim step before transfer to liquid nitrogen.

## Contrôle de la qualité et analyse moléculaire

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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.