

OKT 3 Cells | 300499

General information

Description	Mice were immunized with human peripheral blood lymphocytes. The splenocytes were fused with P3x63Ag8.U1 myeloma cells. The monoclonal antibody produced by this hybridoma was used as an immunosuppressant under the name Muromonab-CD3.
Organism	Mouse
Tissue	Spleen
Applications	Immunology, production of therapeutic antibodies
Synonyms	OKT3

Characteristics

Breed/Subspecies	CAF1 x BALB/c
Morphology	Lymphoblast
Cell type	Hybridoma (Spleen, B cell)
Growth properties	Adherent/Suspension

Regulatory Data

Citation	OKT 3 (Cytion catalog number 300499)
Biosafety level	1
NCBI_TaxID	10090
CellSaurusAccession	CVCL_2665

Biomolecular Data

Products	Immunoglobulin, monoclonal antibody, against human T cell (human T lymphocyte), against CD3
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Handling

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Culture Medium RPMI 1640, w: 2.0 mM stable Glutamine, w: 2.0 g/L NaHCO₃ (Cytion article number 820700a)

Supplements Supplement the medium with 10% heat-inactivated FBS

Subculturing Gently homogenize the cell suspension in the flask by pipetting up and down, then take a representative sample to determine the cell density per ml. Dilute the suspension to achieve a cell concentration of 1×10^5 cells/ml with fresh culture medium, and aliquot the adjusted suspension into new flasks for further cultivation.

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% CO₂, humidified atmosphere.

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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.