RF/6A

305150

GENERAL INFORMATION

Description

RF/6A is a rhesus macaque (Macaca mulatta) retinal choroidal endothelial cell line established from fetal choroid and retina tissue. The line is grows as an adherent monolayer with epithelial-like morphology. RF/6A cells retain key endothelial characteristics including expression of Factor VIII (von Willebrand factor), fibronectin, and Weibel-Palade granules detectable by electron microscopy — the latter confirming their endothelial identity. The line was originally established for studies of retinal and choroidal vascularization and has been widely adopted as a primate endothelial model for ocular angiogenesis research.
RF/6A is applicable in ocular angiogenesis research, studies of retinal and choroidal vascularization, evaluation of anti-angiogenic agents (VEGF inhibitors, bevacizumab, ranibizumab), age-related macular degeneration (AMD) modeling, diabetic retinopathy biology, and assessment of vascular permeability in the ocular microenvironment. The non-human primate (NHP) origin makes RF/6A closer to human retinal vascular biology than rodent endothelial models, particularly for studies involving primate-specific VEGF isoform responses and ocular pharmacology. The line is commonly used in tube formation assays, migration assays, and VEGF-stimulation experiments.

Tissue	Choroid, retina
Disease	Normal retinal choroidal endothelium (fetal; non-tumorigenic)
Metastatic site	Not applicable (normal fetal retinal choroidal endothelial cell line)

Applications

Ocular angiogenesis research; retinal and choroidal vascularization; anti-VEGF therapy evaluation (bevacizumab, ranibizumab); AMD and diabetic retinopathy modeling; tube formation assays; vascular permeability; NHP primate retinal endothelial model

CLASSIFICATION

Organism	Rhesus macaque
Sex	Unspecified
Growth properties	Adherent
GMO status	Non-GMO

GMO status details

No genetic modification; wildtype rhesus macaque fetal retinal choroidal endothelial cell line

Biosafety level	1

CHARACTERISTICS

Morphology	Epithelial-like
Cell type	Endothelial cells
Doubling time	approx. 24 to 36 hours

IDENTIFIERS

Citation

RF/6A (Cytion catalog number 305150)

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NCBI TaxID	9544
Cellosaurus	CVCL_4552

EXPRESSION AND SUSCEPTIBILITY

Protein expression

Factor Ⅷ, Fibronectin

HANDLING AND CULTURE

Culture medium

EMEM (MEM Eagle), w: 2 mM L-Glutamine, w: 2.2 g/L NaHCO3, w: EBSS (Cytion article number 820100a)

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

Supplements

Supplement the medium with 10% FBS and 1% NEAA

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.

Split ratio	1 to 5
Seeding density	1 to 2 × 10^4 cells/cm^2
Fluid renewal	2 to 3 times per week
Post-thaw recovery	After thawing, plate the cells at 5 × 10^4 cells/cm^2 and allow at least 24 hours for adherence before the first medium change. Do not allow cultures to reach full confluency as contact inhibition may reduce endothelial phenotype.

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

Donor age	Fetus
Ethnicity	Not applicable (non-human primate cell line; Macaca mulatta)

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