Product sheet

Cytion

NRK-Pom121-EGFP3 Cells | 500669

General information

Description	The NRK-Pom121-EGFP3 cell line is derived from normal rat kidney (NRK) cells and is genetically engineered to express the Pom121-EGFP3 fusion protein. Pom121 is a transmembrane nucleoporin that is an integral component of the nuclear pore complex (NPC), playing a crucial role in nuclear envelope assembly and NPC function. The inclusion of the enhanced green fluorescent protein (EGFP3) tag facilitates the visualization and study of Pom121 dynamics, localization, and interactions within live cells through fluorescence microscopy. This makes the NRK-Pom121-EGFP3 cell line a valuable tool for investigating nuclear transport mechanisms and NPC architecture. NRK cells, the parental line of NRK-Pom121-EGFP3, are commonly used in various research applications due to their stable growth characteristics and epithelial morphology. The modification to express Pom121-EGFP3 provides researchers with a robust model to examine the molecular mechanisms underlying nucleocytoplasmic transport, the structural organization of the NPC, and its regulation during cell division and differentiation. Additionally, this cell line can be used to study the effects of various genetic and pharmacological perturbations on NPC function, offering insights into diseases associated with nuclear transport defects, such as cancer and neurodegenerative disorders. Overall, the NRK-Pom121-EGFP3 cell line represents a sophisticated tool in cell biology and molecular research, providing high-resolution insights into the dynamic processes governing nucleocytoplasmic interactions. Its ability to allow real-time observation of NPC components in a live cellular context makes it invaluable for advancing our understanding of cellular transport mechanisms and their implications in health and disease.
Organism	Rat
Tissue	Kidney
Synonyms	NRK Pom121-EGFP3, NRK Pom121-3EGFP, NRK-Pom121-3EGFP

Characteristics

Breed/Subspecies	OsborneMendel
Morphology	Fibroblast-like cells with fusiform shape
Growth properties	Monolayer, adherent

Regulatory Data

Citation	NRK-Pom121-EGFP3 (Cytion catalog number 500669)
Biosafety level	1

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NCBI_TaxID	10116
CellosaurusAccession	CVCL_AV96
Depositor	The Ellenberg Lab (EMBL)

Biomolecular Data

Receptors expressed	Epidermal growth factor (EGF), multiplication stimulating activity (MSA)
Protein expression	Pom121-EGFP3: Location/Gene: 1..589 / Pcmv, 653..4250 / Pom121, 4251..4287 / null, 4318..6546 / 3EGFP, 7780..8574 / KanR/NeoR
Products	Epidermal growth factor (EGF), multiplication stimulating activity (MSA), POM121, Transmembrane, Nucleoporin, CMV Promotor, Neomycin, Phosphotransferase

Handling

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)
Supplements	Supplement the medium with 10% FBS, 0.5 mg/mL G418
Dissociation Reagent	Accutase
Subculturing	Discard the old medium and wash the cells with PBS. Add a freshly prepared 0.025% trypsin/0.02% EDTA solution heated to 37 degrees Celsius and wait until the cells detach, which usually takes about 5 minutes. Neutralize the trypsin by adding fresh medium, then transfer the cell mixture to a tube and centrifuge. After centrifugation, remove the supernatant, resuspend the cell pellet in fresh culture medium, and transfer the suspension to new flasks. Incorporate G418 into the culture medium to achieve a final concentration of 0.5 mg/ml
Seeding density	2 to 4 x 10^4 cells/cm^2
Fluid renewal	2 to 3 times per week
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°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_2, 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

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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.