NRK-EGFP2-Nup50

500726

GENERAL INFORMATION

Description

The NRK-EGFP2-Nup50 cell line is a clonal stable cell line derived from normal rat kidney (NRK) cells. This cell line was generated through the transfection of a circular plasmid containing the gene encoding the fusion protein of Enhanced Green Fluorescent Protein (EGFP) and Nucleoporin 50 (Nup50), followed by drug resistance selection. As a result, approximately 50% of the cells express the EGFP3-Nup50 fusion protein, which enables the visualization and tracking of Nup50 within the cellular environment.
Nup50 is a critical component of the nuclear pore complex, which is responsible for regulating the transport of molecules between the nucleus and cytoplasm. The EGFP3 tag allows for live-cell imaging and other fluorescence-based techniques to study the localization, dynamics, and interactions of Nup50. Despite being a stable cell line, the NRK-EGFP2-Nup50 cells exhibit some variegation, indicating variability in the expression levels of the EGFP3-Nup50 fusion protein among the cells.
This cell line is particularly valuable for research focused on nucleocytoplasmic transport, nuclear pore complex dynamics, and the functional role of Nup50 in various cellular processes. The NRK-EGFP2-Nup50 cells are suitable for a range of experimental approaches, including fluorescence recovery after photobleaching (FRAP), fluorescence correlation spectroscopy (FCS), and other advanced microscopy techniques. These studies can provide insights into the molecular mechanisms of nuclear transport and contribute to the understanding of diseases associated with nuclear transport dysfunction, such as certain cancers and neurodegenerative disorders.

Tissue	Kidney

Synonyms

- NRK EGFP2-Nup50

CLASSIFICATION

Organism	Rat
Growth properties	Adherent
Biosafety level	1

CHARACTERISTICS

Morphology	Fibroblast-like cells with fusiform shape

IDENTIFIERS

Citation

NRK-EGFP2-Nup50 (Cytion catalog number 500726)

NCBI TaxID	10116
Cellosaurus	CVCL_AV93
Depositor	The Ellenberg Lab (EMBL)

EXPRESSION AND SUSCEPTIBILITY

Receptors expressed

Epidermal growth factor (EGF), multiplication stimulating activity (MSA)

Cytion GmbH
Nikola-Tesla-Str. 3
69124 Heidelberg
Tel: +49 (0)6221 405780
info@cytion.com
cytion.com

Registered office Heidelberg
VAT ID: DE283292917
Local court Mannheim - HRB 714394
CEO / Geschäftsführer: Jonathan Steubing

Banking information
Sparkasse Heidelberg
IBAN: DE35 6725 0020 0009 2076 86
BIC: SOLADES1HDB

Protein expression

EGFP3-Nup50

Products

NUP50 (Nucleoporin 50)

HANDLING AND CULTURE

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)

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

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

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

Breed / subspecies	OsborneMendel

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.

Disclaimer

Our cells are provided for in vitro laboratory research purposes exclusively and are not intended for clinical or diagnostic use, nor are they to be administered to humans or used for veterinary purposes. Users must adhere to all applicable guidelines and regulations for the handling and use of these cells in a research setting.

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.

Third-party agreements apply	Yes