Product sheet

NRK-Pom121-EGFP3 growing culture | 550669

General information

Description	This clonal stable cell line was generated by transfection of a circular plasmid (see below) followed by drug resistance selection. Add G418 to culture medium at a final concentration of 0.5 mg/ml.
Organism	Rat
Tissue	Kidney
Synonyms	NRK Pom121-EGFP3, NRK Pom121-3EGFP, NRK-Pom121-3EGFP

Characteristics

Morphology	Fibroblast-like cells with fusiform shape
Growth properties	Monolayer, adherent

Identifiers / Biosafety / Citation

Citation	NRK-Pom121-EGFP3 (Cytion catalog number 500669)
Biosafety level	1
Depositor	Dr. J. Ellenberg, EMBL Heidelberg

Expression / Mutation

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

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Medium supplements	10% FBS, w: 4.5 g/L Glucose, w: 4 mM L-Glutamine, w: 1.5 g/L NaHCO3, w: 1.0 mM Sodium pyruvate, Add G418 to culture medium at a final concentration of 0.5 mg/ml
Passaging solution	Accutase
Subculturing	Remove medium and rinse with PBS. Add fresh 0.025% trypsin/0.02% EDTA solution at 37 degree Celsius until cells detach (typically ~5 min). To remove trypsin, add fresh medium, transfer to a tube and centrifuge. Aspirate the supernatant, resuspend the cell pellet in culture medium and dispense into new flasks. Add G418 to culture medium at a final concentration of 0.5 mg/ml.
Split ratio	A ratio of 1:3 to 1:4 is recommended
Seeding density	2 to 4 x 10^4 cells/cm^2
Fluid renewal	2 to 3 times per week
Freeze medium	CM-1 (Cytion catalog number 800100) or CM-ACF (Cytion catalog number 806100)
Handling of cryopreserved cultures	The cells come deep-frozen shipped on dry ice. Please make sure that the vial is still frozen. If immediate culturing is not intended, the cryovial must be stored below -150 degree Celsius after arrival. If immediate culturing is intended, please follow the below instructions: Quickly thaw by rapid agitation in a 37 degree Celsius water bath within 40-60 seconds. The water bath should have clean water containing an antimicrobial agent. As soon as the sample has thawed, remove the cryovial from the water bath. A small ice clump should still remain and the vial should still be cold. From now on, all operations should be carried out under aseptic conditions. Transfer the cryovial to a sterile flow cabinet and wipe with 70% alcohol. Carefully open the vial and transfer the cell suspension into a 15 ml centrifuge tube containing 8 ml of culture medium (room temperature). Resuspend the cells carefully. Centrifuge at 300 x g for 3 min and discard the supernatant. The centrifugation step may be omitted, but in this case the remains of the freeze medium have to be removed 24 hours later. Resuspend the cells carefully in 10 ml fresh cell culture medium and transfer them into two T25 cell culture flasks. All further steps are described in the subculture section.
Handling of proliferating cultures	One or two cell culture flasks come filled with cell culture medium. Collect the entire medium in 1 or 2 x 50 ml centrifuge tubes, respectively. Carefully add 5 ml of cell culture medium to each T25 cell culture flask. Control the cell morphology and confluency under the microscope. Incubate at 37 degree Celsius for a minimum of 24 hours. Spin down the collected medium at 300 x g for 3 minutes to collect the cells which may have detached during transit. If a cell pellet is visible, resuspend the cells in 5 ml of cell culture medium and transfer to a T25 cell culture flask. Incubate at 37 degree Celsius for a minimum of 24 hours.

Quality control / Genetic profile / HLA

Sterility	Mycoplasma contaminations are ruled out through PCR-based and luminescence-based mycoplasma assays. The absence of bacterial, fungal or yeast contamination is controlled through daily visual cell monitoring.

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STR profile

Rat_D1Wox31: 96,100
Rat_D2Wox37: 156
Rat_D19Wox11: 220
Rat_D10Wox8: 266,270
Rat_D4Wox7: 153,157
Rat_D2Wox27: 211
Rat_D5Rat33: 116,138
Rat_D10Wox11: 156
Rat_D1Wox23: 210,214
Rat_D12Wox1: 402,406
Rat_D6Wox2: 104,124
Rat_D8Wox7: 185
Rat_D6Cebr1: 221,233
SRY: x,Y