

## NCH612 Cells | 300121

### General information

#### Description

NCH612 is a patient-derived oligodendrocytic cell line that originates from human brain tissue and serves as a relevant research model for anaplastic oligodendroglioma (WHO grade III). This cell line harbors the IDH1 R132H mutation, a hallmark genetic alteration frequently associated with oligodendrogliomas. The mutation leads to epigenetic modifications, including the glioma CpG island methylator phenotype (G-CIMP), which contributes to tumor development and progression. Notably, NCH612 exhibits a partial deletion of chromosome arms 1p and 19q, a genetic characteristic commonly found in oligodendrogliomas and associated with better prognosis and response to certain therapies.

Studies have demonstrated that NCH612 is particularly sensitive to the DNA methyltransferase inhibitor decitabine (DAC). Treatment with DAC results in reduced cell proliferation and colony formation, primarily through the downregulation of TERT (telomerase reverse transcriptase) and the upregulation of p21, a cyclin-dependent kinase inhibitor involved in the DNA damage response. Interestingly, this sensitivity appears to be linked to the presence of both the IDH1 mutation and 1p/19q codeletion, as other IDH1-mutant glioma cell lines without this deletion, such as NCH1681, exhibit resistance to DAC. These findings suggest that epigenetic therapies like DAC could be particularly effective in IDH1-mutant anaplastic oligodendrogliomas with 1p/19q codeletion.

Further molecular investigations reveal that DAC treatment in NCH612 cells leads to the enrichment of pathways related to DNA replication, cell cycle regulation, and lysosomal function, shedding light on the drug's mechanism of action. The repression of TERT by DAC is mediated by p21, emphasizing the critical role of this pathway in the response to epigenetic therapy. Given its well-defined genetic and epigenetic profile, NCH612 represents a valuable in vitro model for studying the biology of anaplastic oligodendrogliomas and for developing targeted therapies aimed at IDH1-mutant tumors with 1p/19q codeletion.

**Organism** Human

**Tissue** Brain

**Disease** Anaplastic oligodendroglioma, WHO grade III, IDH1 mutant (R132H)

### Characteristics

**Age** 39 years

**Gender** Male

**Ethnicity** Caucasian

**Growth properties** Spheroid culture

### Regulatory Data

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<b>Citation</b>	NCH612 (Cytion catalog number 300121)
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<b>Biosafety level</b>	1
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<b>NCBI_TaxID</b>	9606
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<b>CellosaurusAccession</b>	CVCL_x913
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## Biomolecular Data

## Handling

<b>Culture Medium</b>	DMEM:Ham's F12 (1:1), w: 3.1 g/L Glucose, w: 2.5 mM L-Glutamine, w: 15 mM HEPES, w: 0.5 mM Sodium pyruvate, w: 1.2 g/L NaHCO <sub>3</sub> (Cytion article number 820400a)
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<b>Supplements</b>	Supplement the medium with 10% FBS, 5 mg/L Heparin, 20 ng/mL bFGF, 20 microgram/L EGF, 5 mg/L Insulin, 100 mg/L Transferrin, 5,2 microgram/L Na-selenit, 6,3 microgram/L Progesteron, 161,1 microgram/L Putrescin, 50 mg/L Hydrocortinson
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<b>Subculturing</b>	For subculturing spheroid cultures, begin by mechanically dissociating the spheroids through pipetting up and down 5 to 10 times using an Eppendorf pipette with 1000 µl filter tips. After this, centrifuge the mixture at 300g for 5 minutes at room temperature to pellet the cells. Discard the supernatant and resuspend the cell pellet in fresh culture medium. Finally, transfer the resuspended cells into new culture vessels to promote further spheroid formation. This approach ensures efficient spheroid breakdown and readies them for continued growth in a new environment.
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<b>Seeding density</b>	1 x 10 <sup>5</sup> cells/mL
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<b>Fluid renewal</b>	Fresh medium must be added every 2to3 days (2to5 ml depending on the size of the cell culture flask).
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<b>Post-Thaw Recovery</b>	Slow. After thawing allow the cells to recover from the freezing process for at least 48 hours.
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<b>Freeze medium</b>	As a cryopreservation medium, we use 50% basal medium + 40% FBS + 10% DMSO, 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  $-150^{\circ}\text{C}$  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  $37^{\circ}\text{C}$  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  $300 \times g$  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

$37^{\circ}\text{C}$ , 5%  $\text{CO}_2$ , humidified atmosphere.

### Shipping Conditions

Cryopreserved cell lines are shipped on dry ice in validated, insulated packaging with sufficient refrigerant to maintain approximately  $-78^{\circ}\text{C}$  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  $-196^{\circ}\text{C}$ . Storage at  $-80^{\circ}\text{C}$  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.