

## C6 Cells | 500142

## General information

## Description

The C6 cell line maintains glial cell type with fibroblast morphology and originates from a glioma of a Wistar-Furth rat. The glioma was induced by exposure to N-nitrosomethylurea, following numerous cycles of alternating culture and animal passages.

The C6 glioma cell line is frequently utilized in neuro-oncology research to create animal models that closely mimic the characteristics of human glioma, aiding in the development of new therapeutic agents and strategies. It is particularly effective in 3D cell culture and high-throughput screening.

C6 cells are genetically diverse, possessing a wild-type p53 gene, increased Rb gene expression, and a mutant p16/Cdkn2a/Ink4a locus but lacking p16 and p19ARF mRNA expression. They also overexpress several genes in human gliomas, such as PDGF $\beta$ , IGF-1, EGFR, and Erb3/Her3 precursor proteins.

However, the expression of IGF-2, FGF-9, and FGF-10 is reduced, while MMP-7 gene expression remains unchanged. Like human gliomas, C6 cells show increased activity of the Ras pathway genes, which is regulated by the elevated expression of the Ras guanine triphosphate activator protein.

The C6 cell line has been utilized in various studies. For instance, it was used to examine the ability of 2-(2,4-dihydroxy phenyl)thieno-1,3-thiazin-4-one (BChTT) to halt cancer cell proliferation and to investigate the mechanisms involved in this process.

In another research, the cytotoxic and antioxidant properties of the supercritical CO<sub>2</sub> extract (SCE) of an old man's beard (*Usnea barbata*) were studied using C6 cells. Interestingly, these cells have been reported to show increased levels of glycerol phosphate dehydrogenase activity in response to glucocorticoids.

**Organism** Rat

**Tissue** Brain

**Disease** Glioma

**Synonyms** C-6, C 6, RGC-6, RGC6, RGC6

## Characteristics

**Age** Unspecified

**Gender** Male

**Morphology** Fibroblast-like

**Cell type** Glial cells

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**Growth properties** Adherent

### Identifiers / Biosafety / Citation

**Citation** C6 (Cytion catalog number 500142)

**Biosafety level** 1

### Expression / Mutation

**Receptors expressed** Glucocorticoid

**Viruses** Positive for LCMV

**Virus susceptibility** vesicular stomatitis (Indiana), vaccinia, herpes simplex

**Virus resistance** poliovirus 3

**Reverse transcriptase** negative

**Products** S-100 protein, production of glyceryl phosphate dehydrogenase in response to glucocorticoids, somatotrophin.

### Handling

**Culture Medium** RPMI 1640, w: 2.1 mM stable Glutamine, w: 2.0 g/L NaHCO<sub>3</sub> (Cytion article number 820700a)

**Medium supplements** Supplement the medium with 10% FBS

**Passaging solution** Accutase

**Doubling time** 24 hours

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**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** A ratio of 1:2 to 1:3 is recommended

**Seeding density**  $1 \times 10^4$  cells/cm<sup>2</sup> will yield in a confluent layer in about 4 days

**Fluid renewal** 2 to 3 times per week

**Freezing recovery** After thawing, plate the cells at  $5 \times 10^4$  cells/cm<sup>2</sup> and allow the cells to recover from the freezing process and to adhere for at least 24 hours.

**Freeze medium** CM-1 (Cytion catalog number 800100) or CM-ACF (Cytion catalog number 806100)

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### Handling of cryopreserved cultures

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. Optionally, skip centrifugation but remove any remaining freezing medium after 24 hours.
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.

## Quality control / Genetic profile / HLA

### 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.

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**STR profile**

**Rat\_D1Wox31:** 104  
**Rat\_D2Wox37:** 156  
**Rat\_D19Wox11:** 220,228  
**Rat\_D10Wox8:** 266  
**Rat\_D4Wox7:** 145  
**Rat\_D2Wox27:** 207,215  
**Rat\_D5Rat33:** 122  
**Rat\_D10Wox11:** 156,171  
**Rat\_D1Wox23:** 214  
**Rat\_D12Wox1:** 406  
**Rat\_D6Wox2:** 104  
**Rat\_D8Wox7:** 182  
**Rat\_D6Cebr1:** 233,239  
**SRY:** x,Y