

C6 growing culture | 550142

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 PDGFβ, 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 CO2 extract (SCE) of an old man's beard (<i>Usnea barbata</i>) were studied using C6 cells. Interestingly, these cells have been reported to show increased levels of glyceryl 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
Growth properties	Adherent

Identifiers / Biosafety / Citation

Citation	C6 (Cytion catalog number 500142)
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Biosafety level 1

Expression / Mutation

Receptors expressed	Glucocorticoid
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
Medium supplements	10% FBS, w: 2.1 mM stable Glutamine, w: 2.0 g/L NaHCO ₃
Passaging solution	Accutase
Doubling time	24 hours
Subculturing	Remove medium and rinse the adherent cells using PBS without calcium and magnesium (3-5 ml PBS for T25, 5-10 ml for T75 cell culture flasks). Add Accutase (1-2 ml per T25, 2.5 ml per T75 cell culture flask), the cell sheet must be covered completely. Incubate at ambient temperature for 8-10 minutes. Carefully resuspend the cells with medium (10 ml), centrifuge for 3 min at 300 g, resuspend cells in fresh medium and dispense into new flasks which contain fresh medium.
Split ratio	A ratio of 1:2 to 1:3 is recommended
Seeding density	1 x 10 ⁴ cells/cm ² will yield in a confluent layer in about 4 days
Fluid renewal	2 to 3 times per week

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Freezing recovery

After thawing, plate the cells at 5×10^4 cells/cm² 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)

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.

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