Product sheet

Cytion

C2C12 Cells | 400476

Description	The C2C12 cell line, an immortalized mouse myoblast cell line derived from the thigh muscle of a 2-month-old mouse of the C3H mouse strain, is extensively utilized in biomedical research for its unique cell differentiation properties. C2C12 myoblast cells proliferate rapidly and exhibit typical myoblast characteristics under high serum conditions. Upon shifting to low serum conditions or starvation, C2C12 cells initiate myogenic differentiation, transitioning into myotubes, which are precursors to contractile skeletal muscle cells. C2C12 cells incorporate exogenous cDNA and nucleic acids through transfection easily, making them a good choice for gene expression studies and investigations into myoblasts and myotubes differentiation. The differentiation process is marked by the expression of myogenic markers such as Myf5, MyoD, Myogenin, and Mrf4, alongside muscle-specific markers like Csrp3 and Mef2a, which are essential in studying different muscle phenotypes and skeletal muscle regeneration. The unique shape of C2C12 myoblasts and their transformation into myoblast cell rings and subsequently into mature myotubes in serum-supplemented media underscore the dynamic nature of these cells and their potential in skeletal muscle research. Researchers use substrates like gelatin hydrogels for C2C12 cell cultures to simulate in vivo muscle conditions, enabling detailed studies of muscle cell development and extracellular matrix effects. Metabolic profiling reveals key insights into the pathways involved in muscle formation and recovery, focusing on essential proteins and calcium's role in contraction. Gene silencing techniques further illuminate the differentiation process, highlighting the significance of SMAD1 phosphorylation in muscle regeneration, crucial for understanding recovery in muscle wasting and injury. In summary, the C2C12 cell line serves as a critical tool in the realm of biomedical research, offering a versatile platform for exploring the intricacies of muscle formation, differentiation, gene expression, and the profound impact of various factors on the skeletal muscle cell lineage, including the pivotal role of myofilaments, intermediate filament proteins, and the overall organismal context in which these cellular processes unfold.
Organism	Mouse
Tissue	Muscle
Applications	Transfection host
Synonyms	C2c12, C2-C12, C12

Breed/Subspecies	C3H
Age	2 months
Gender	Female

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Morphology	Myoblast-like
Cell type	Myoblast
Growth properties	Adherent

Citation	C2C12 (Cytion catalog number 400476)
Biosafety level	1
NCBI_TaxID	10090
CellosaurusAccession	CVCL_0188

Culture Medium	RPMI 1640, w: 2.0 mM stable Glutamine, w: 2.0 g/L NaHCO3 (Cytion article number 820700a)
Supplements	Supplement the medium with 10% FBS
Dissociation Reagent	Accutase
Doubling time	24 hours
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.
Seeding density	1 x 10^4 cells/cm^2 will yield in a confluent layer in about 4 days
Fluid renewal	Every 3 to 5 days

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Post-Thaw Recovery	After thawing, plate the cells at 5×10^4 cells/cm^2 and allow the cells to recover from the freezing process and to adhere for at least 24 hours.
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.
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°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°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 x 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°C, 5% 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 °C throughout transit. On receipt, inspect the container immediately and transfer vials without delay to appropriate storage.

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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.

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.