Cell line profile

C2C12 (ECACC catalogue no. 91031101)

Cell line history
C2C12 is a subclone from a myoblast line established from normal adult C3H mouse leg muscle\(^1\). The original cell line, C2, was obtained by Yaffe and Saxel in 1977 by establishing primary cultures from the thigh muscle of 2 month old normal mice, 70 hours after crush injury\(^2\).

Key characteristics
Differentiates rapidly; produces extensive contracting myotubes, expressing characteristic muscle proteins. Recombinant bone morphogenetic protein-2 (BMP-2) has been shown to inhibit the formation of multinucleated myotubes formed by confluent C2C12 cells, which in turn leads to the shift of the differentiation pathway of the cells from a myoblastic lineage to an osteoblastic lineage\(^3\).

Applications
C2C12 is one of the most popular cell lines supplied by ECACC. These cells provide in vitro models to study differentiation of myoblasts and osteoblasts and mechanistic biochemical pathways. C2C12 cells have also been used to aid the understanding of early myogenesis by identifying the underlying regulatory network performed by transcriptional regulators, using profiling and shRNA screens\(^4\). Due to their high division rate, C2C12 cells are useful in studying the cell cycle. For example, the mechanism of X inactivation, via the highly coordinated replication of Xi in early S-phase of the cell cycle, was studied in C2C12 cells using GFP-tagged replication proteins and BrdU-labelled nucleotides\(^5\).

Culture tips
C2C12 cells should be cultured in DMEM + 2mM Glutamine + 10-15% Foetal Bovine Serum (FBS). Cells can be relatively slow growing when resuscitated from frozen taking 4-5 days to

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24 hours post resuscitation

Prior to cryopreservation
reach 50% confluence when seeded at 2x1,000 cells/cm². Split semi-
confluent cultures (50 - 70%) 1:3 to 1:6 i.e. seeding at 1-2x1,000 cells/cm² using 0.05%
trypsin or trypsin/EDTA; 5% CO2; 37°C. Do not allow cells to reach confluence as myotube
formation may occur.

Key references
[2] Yaffe D and Saxel O. Serial passaging and differentiation of myogenic cells isolated from
expressed transcriptional regulators whose knock-down inhibits myoblast differentiation.
[5] Casas-Delucchi C et al. Histone acetylation controls the inactive X chromosome

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