**Product sheet**

# MC3T3-E1 Cells | 305187

## General information

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| **Description** | MC3T3-E1 is a pre-osteoblastic cell line derived from the calvaria of a mouse embryo. These cells are extensively utilized in the study of osteogenesis, particularly for examining the molecular and cellular mechanisms underlying bone formation and differentiation. The MC3T3-E1 cell line is known for its robust ability to differentiate into osteoblasts in vitro, a process that can be stimulated by ascorbic acid and beta-glycerophosphate. This differentiation is marked by the expression of key osteogenic markers such as alkaline phosphatase, osteocalcin, and type I collagen.<br><br>MC3T3-E1 cells are instrumental in research focused on bone biology, including the study of bone matrix deposition and mineralization. These cells provide a reliable model for investigating the effects of various drugs, hormones, and genetic modifications on osteoblast function and bone formation. Additionally, the MC3T3-E1 cell line is valuable in studying pathological conditions such as osteoporosis and other bone-related diseases. Their ease of culture and well-characterized response to osteogenic stimuli make them a preferred choice for researchers aiming to unravel the complexities of bone physiology and pathology. |
| **Organism** | Mouse |
| **Tissue** | Bone, calvaria |
| **Applications** | In vitro osteoblast differentiation |
| **Synonyms** | Mc3T3-E1, MC3T3E1, MC-3T3-E1, MC 3T3-E1 |

## Characteristics

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| **Breed/Subspecies** | C57BL/6 |
| **Age** | 1 day |
| **Gender** | Unspecified |
| **Morphology** | Fibroblast-like |
| **Cell type** | Osteoblast |
| **Growth properties** | Adherent |

## Regulatory Data

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| **Citation** | MC3T3-E1 (Cytion catalog number 305187) |

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| **Biosafety level** | 1 |
| **NCBI_TaxID** | 10090 |
| **CellosaurusAccession** | CVCL_0409 |

## Biomolecular Data

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| **Tumorigenic** | Yes, in immunodeficient mice |
| **Products** | Collagen |

## Handling

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| **Culture Medium** | Alpha MEM, w: 2.0 mM stable Glutamine, w: Ribonucleosides, w: Deoxyribonucleosides, w: 1.0 mM Sodium pyruvate, w: 2.2g/L NaHCO3, w/o: Ascorbic acid (GIBCO, Catalog No. A1049001. We do not supply this product; please consider other suppliers. Please let us know if you need further assistance.) |
| **Supplements** | Supplement the medium with 10% FBS |
| **Dissociation Reagent** | Accutase |
| **Doubling time** | 24 to 48 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. |
| **Fluid renewal** | 2 to 3 times per week |
| **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. |

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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.<br>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.<br>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.<br>4. Perform all subsequent steps under sterile conditions in a flow hood, disinfecting the cryovial with 70% ethanol before opening.<br>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.<br>6. Centrifuge the mixture at 300 x g for 3 minutes to separate the cells and carefully discard the supernatant containing residual freezing medium.<br>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.<br>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. |
| **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. |

## Quality Control & Molecular Analysis

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| **Sterility** | Mycoplasma contamination is excluded using both PCR-based assays and luminescence-based mycoplasma detection methods.<br><br>To ensure there is no bacterial, fungal, or yeast contamination, cell cultures are subjected to daily visual inspections. |