Product sheet

NE-4C Cells | 305182

General information

Description	NE-4C cells are a murine neuroectodermal stem cell line derived from the cerebral vesicles of embryonic mouse brain tissue. The cell line is widely recognized for its capacity to undergo neuronal differentiation under defined experimental conditions, making it a valuable in vitro model for studies of neurogenesis, neural development, and neuronal lineage commitment. NE-4C cells exhibit characteristics of neural progenitor cells and can be induced to differentiate into neuron-like cells through treatment with agents such as retinoic acid, resulting in morphological and molecular changes associated with neuronal maturation. During differentiation, NE-4C cells develop extended neurite-like processes and express neuronal markers including βIII-tubulin, MAP2, and neurofilament proteins, depending on the differentiation protocol employed. Because of their relatively homogeneous differentiation behavior and reproducible response to inductive stimuli, NE-4C cells are commonly used to investigate signaling pathways involved in neural specification, synaptic development, neurotoxicity, oxidative stress responses, and neuroprotective mechanisms. The model has also been applied in studies of neurodegenerative disease mechanisms, developmental neurobiology, and screening of compounds affecting neuronal differentiation or survival.
Organism	Mouse
Tissue	Brain, Neuroectodermal

Characteristics

Breed/Subspecies	C57BL/6 x 129/Sv-Trp53-/-
Age	Embryo
Morphology	Neuroepithelial
Growth properties	Adherent

Regulatory Data

Citation	NE-4C (Cytion catalog number 305182)
Biosafety level	1
NCBI_TaxID	10090
CellosaurusAccession	CVCL_B063

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Biomolecular Data

Antigen expression	Sox-2, Otx-2, En-1

Handling

Culture Medium	EMEM (MEM Eagle), w: 2 mM L-Glutamine, w: 2.2 g/L NaHCO3, w: EBSS (Cytion article number 820100a)
Supplements	Supplement the medium with 10% FBS and 1% NEAA
Dissociation Reagent	Accutase
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 to 3 x 10^4 cells/cm^2
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.

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.
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

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.