Product sheet

Ku 80-/- Cells | 305258

General information

Description	Ku80-/- MEF (Mouse Embryonic Fibroblast) cells are genetically engineered fibroblast cells derived from mice that lack the Ku80 gene (XRCC5). The Ku80 protein, in conjunction with Ku70, forms the Ku heterodimer, which is essential for the non-homologous end joining (NHEJ) pathway of DNA double-strand break (DSB) repair. The absence of Ku80 in these cells impairs their ability to effectively repair DSBs, making them a valuable model for studying the role of the NHEJ pathway in genomic stability, DNA repair mechanisms, and cancer biology. Ku80-/- MEF cells exhibit increased sensitivity to ionizing radiation and other DNA-damaging agents due to their compromised DSB repair capacity. These cells also tend to accumulate chromosomal aberrations and exhibit genomic instability. The lack of Ku80 affects not only DNA repair but also other cellular processes such as V(D)J recombination, which is crucial for the development of a diverse repertoire of antibodies and T-cell receptors in the immune system. Research using Ku80-/- MEF cells has provided significant insights into the molecular mechanisms of NHEJ and the broader implications of defective DNA repair. These studies are crucial for understanding the development of cancer and other diseases associated with genomic instability. Additionally, they help in the exploration of potential therapeutic targets for enhancing DNA repair in cancer cells, thereby improving the efficacy of cancer treatments that rely on inducing DNA damage in tumor cells.
Organism	Mouse
Tissue	Embryo
Disease	Normal mouse embryonic fibroblast (Ku80/XRCC5 knockout; SV40-immortalized; NHEJ-deficient)
Metastatic site	Not applicable (immortalized MEF; not a clinical tumor sample)
Applications	NHEJ DNA repair research; Ku80/XRCC5 function; DNA double-strand break (DSB) repair; genomic instability modeling; radiation sensitivity; V(D)J recombination; cancer biology; genotoxicity testing; DNA damage response
Synonyms	Ku80-/- MEF

Characteristics

Age	12-13 fetal days
Gender	Unspecified
Ethnicity	Not applicable (mouse cell line)
Morphology	Fibroblast-like

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Cell type	Fibroblast
Growth properties	Adherent

Regulatory Data

Citation	Ku 80-/- (Cytion catalog number 305258)
Biosafety level	2
NCBI_TaxID	10090
CellosaurusAccession	CVCL_UJ16
GMO Status	GMO-S1: This MEF line carries homozygous Ku80 (XRCC5) knockout and SV40 large T antigen immortalization. The SV40 transgene is stably integrated. This classification applies only within Germany and may differ elsewhere.

Biomolecular Data

Viruses	Transformant: Simian virus 40 (SV40)
Mutational profile	Mutation: Ku80-/-

Handling

Culture Medium	DMEM, w: 4.5 g/L Glucose, w: 4 mM L-Glutamine, w: 3.7 g/L NaHCO3, w: 1.0 mM Sodium pyruvate (Cytion article number 820300a)
Supplements	Supplement the medium with 10% FBS
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.

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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.