

## E11 Cells | 400494

### General information

#### Description

The E11 cell line is a highly specialized murine cell line developed for advanced studies in podocyte function and kidney disease mechanisms. Derived from the glomeruli of transgenic mice engineered to express a temperature-sensitive variant of the SV40 large T antigen, the E11 cells operate under the regulation of the IFN- $\gamma$ -inducible H-2kb promoter. This unique genetic framework facilitates the conditional proliferation of the cells, dependent on the environmental temperature, which aligns with the controlled expression of the T antigen.

One of the distinguishing features of the E11 cell line is its phenotypic stability across extensive passaging. Maintaining consistent expression and cellular characteristics through more than 40 passages, E11 cells have proven invaluable for long-term studies without the common issue of phenotypic drift seen in many cultured cell lines. This stability enhances their use in repetitive and extended biological experiments requiring consistent cell behavior.

In terms of protein expression, the E11 cell line exhibits a robust profile that is quintessential for podocyte-specific studies. The cells consistently express nephrin, an essential component of the slit diaphragm structure in podocytes, alongside a variety of other podocyte-specific proteins such as podocin, CD2AP, and synaptopodin. This comprehensive protein expression facilitates the study of podocyte biology in a controlled in vitro environment, closely simulating in vivo conditions. The ability of E11 cells to form extensive cell-cell contacts further underscores their suitability for modeling kidney filtration barrier functionalities.

**Organism** Mouse

**Tissue** Kidney

### Characteristics

**Breed/Subspecies** (CBA/Ca x C57BL/10)Tg(H2KbtsA58)

**Age** Adult

**Gender** Unspecified

**Cell type** Podocyte

**Growth properties** Adherent

### Regulatory Data

**Citation** E11 (Cytion catalog number 400494)

**Biosafety level** 1

**E11 Cells | 400494****NCBI\_TaxID** 10090**CellosaurusAccession** CVCL\_5737**GMO Status** GMO-S1: This Immorto Mouse podocyte line contains a temperature-sensitive SV40 T-antigen construct enabling conditional immortalization. This classification applies only within Germany and may differ elsewhere.**Biomolecular Data****Protein expression** WT1, Lmx1b, nephrin, NEPH1, FAT, P-cadherin, CD2AP, ZO-1, podocalyxin, podoplanin, synpo, podocin, TRPC6 and GAPDH.**Handling****Culture Medium** RPMI 1640, w: 2.0 mM stable Glutamine, w: 2.0 g/L NaHCO<sub>3</sub> (Cytion article number 820700a)**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.**Seeding density** Inoculate T75 cell culture flasks with  $1 \times 10^4$  cells/cm<sup>2</sup> for the proliferation process. Keep the cells at 33 degree Celsius / 5% CO<sub>2</sub>, until the flask is about 75% confluent.**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.
2. Upon receipt, either store the cryovial immediately at temperatures below  $-150^{\circ}\text{C}$  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  $37^{\circ}\text{C}$  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  $300 \times g$  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

$33^{\circ}\text{C}$ , 5%  $\text{CO}_2$ , humidified atmosphere.

### Shipping Conditions

Cryopreserved cell lines are shipped on dry ice in validated, insulated packaging with sufficient refrigerant to maintain approximately  $-78^{\circ}\text{C}$  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  $-196^{\circ}\text{C}$ . Storage at  $-80^{\circ}\text{C}$  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.

To ensure there is no bacterial, fungal, or yeast contamination, cell cultures are subjected to daily visual inspections.