

**2V6.11 Cells | 305147****General information****Description**

2v6.11 cells were derived from the human embryonic kidney line HEK-293 in 2001. The 2V6.11 cell line is a valuable resource for studying the adenoviral E4 oncoprotein, particularly the E4 34K protein known to be involved in cellular genome maintenance and repair. 2V6.11 cells, obtained through transfection with the plasmid pVgRxR followed by pEKORF6, result in the inducible expression of the E4 34K protein, which is linked to the inhibition of cellular mechanisms that repair double strand-breaks in DNA. The 2V6.11 cell line demonstrated that the adenoviral proteins E4 34k and E1b 55k inhibit chromosomal DNA repair by disrupting non-homologous end joining (NHEJ) and destabilizing DNA repair proteins, extending their effect from extrachromosomal to cellular genomic DNA.

The 2V6.11 inducible cell line, with their adherent epithelial morphology, are ideal for investigating the behavior and characteristics of kidney-derived epithelial cells, including their response to infections by human adenovirus 40. This versatile cell line, which can be detected by western blot, enables researchers to delve into the molecular mechanisms by which the adenovirus E4 oncoprotein inhibits repair processes, thus contributing to our understanding of adenovirus pathology and the potential for developing new therapeutic strategies.

**Organism** Human**Tissue** Fetal Kidney**Characteristics****Age** Fetus**Gender** Female**Morphology** Epithelial**Growth properties** Adherent**Regulatory Data****Citation** 2V6.11 (Cytion catalog number 305147)**Biosafety level** 2**NCBI\_TaxID** 9606**CellosaurusAccession** CVCL\_6355

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### GMO Status

GMO-S1: This HEK293-derived line contains an adenovirus 5 E4-34k expression construct controlled by an ecdysone-inducible promoter, enabling regulated E4 protein production. This classification applies only within Germany and may differ elsewhere.

## Biomolecular Data

## Handling

### Culture Medium

EMEM (MEM Eagle), w: 2 mM L-Glutamine, w: 2.2 g/L NaHCO<sub>3</sub>, 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.

### 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

$37^{\circ}\text{C}$ , 5%  $\text{CO}_2$ , humidified atmosphere.

### Flask Coating

None

### 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.