

HEK293 suspension-adapted | 300686**General information****Description**

The HEK293 suspension-adapted cell line is a variant of the human embryonic kidney 293 (HEK293) cells that has been modified to grow in suspension culture rather than adherent culture. This adaptation is significant for industrial applications where large-scale protein production is required. The cells maintain many of the characteristics of the original HEK293 line, including a robust transient transfection efficiency and the ability to post-translationally modify expressed proteins in a manner similar to that of native human cells.

These cells are particularly valued in the biotechnology and pharmaceutical industries for the production of recombinant proteins and viruses for gene therapy and vaccine development. The adaptation to suspension culture allows for easier scalability and simplifies the harvesting process, making it more suitable for commercial-scale bioprocessing. The HEK293 suspension-adapted cell line supports various viral production systems, including adenovirus, lentivirus, and adeno-associated virus (AAV), which are pivotal in therapeutic applications and research.

Overall, the HEK293 suspension-adapted cell line is a crucial tool in the fields of molecular biology and bioprocessing, providing a versatile platform for the production of various biologically active molecules. Its ease of genetic manipulation and ability to produce proteins that are correctly folded and post-translationally modified according to human cell patterns make it an indispensable resource in many advanced therapeutic and research settings.

Organism Human**Tissue** Kidney**Applications** Transfection host**Characteristics****Age** Fetus**Gender** Female**Morphology** Round**Growth properties** Suspension**Regulatory Data****Citation** HEK293 suspension-adapted (Cytion catalog number 300686)**Biosafety level** 1

HEK293 suspension-adapted | 300686**NCBI_TaxID** 9606**CellosaurusAccession** CVCL_0045**GMO Status** GMO-S1: This suspension-adapted HEK293 cell line contains adenovirus 5-derived E1 sequences from the parental HEK293 line, supporting high proliferative and protein expression capacity. The modification is stably present in transformed embryonic kidney cells. This classification applies only within Germany and may differ elsewhere.**Biomolecular Data****Receptors expressed** Vitronectin**Protein expression** CEA negative, p53 positive**Tumorigenic** In nude mice**Virus susceptibility** Transformed with adenovirus 5 DNA adenovirus 5 DNA**Handling****Culture Medium** Panserin 293S (PanBiotech, Germany)**Supplements** No supplements required**Dissociation Reagent** Not required**Subculturing** Maintain the suspension cells at cell densities between 5×10^5 and $2-3 \times 10^6$ cells/ml in Eppendorf cell culture flasks on a shaker inside an incubator at 37°C/5% CO₂. Subculture once the cell density has reached $2-3 \times 10^6$ cells/ml. Carefully dislodge the cells to avoid cluster. Once the cell density of $1-2 \times 10^6$ cells/ml is achieved, collect the cells by centrifuging at 200xg for 5 min and discard the supernatant. Dilute in an appropriate volume of fresh, prewarmed culture medium and count the cells to get information on the viability and number of cells. Collect the cells by centrifuging at 200xg for 5 min and discard the supernatant. Resuspend the cells in the appropriate volume of freeze medium and count once more. The cell viability should be >>80%, a cell density of 5-10 million cells/ml is recommended. Pipette the cells into pre-labeled cryovials. Use either CoolCell freezing container or a controlled rate freezer to ensure a cooling rate of 1°C/min.

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Seeding density 5 x 10⁵ cells/ml

Post-Thaw Recovery Initiate cultures at a density of 5 x 10⁵ cells/ml and keep the cell concentration up to 2-3 x 10⁶ cells/ml for optimal growth. Incubate at 37°C/5% CO₂ on a cell shaker at 100-150 rpm.

Freeze medium As a cryopreservation medium, we use complete growth medium + 10% DMSO for adequate post-thaw viability.

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 200 x g for 5 minutes, carefully discard the supernatant containing freezing medium.
7. Follow the procedure described under Post-Thaw Recovery

Incubation Atmosphere 37°C, 5% CO₂, 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.

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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.