

**EA.hy926 Cells | 305034**

**Description**

EA.hy926 cells, are a somatic hybrid cell line widely used in cardiovascular disease research. They are employed in studying various aspects of endothelial cell functions related to angiogenesis, homeostasis/thrombosis, blood pressure regulation, and inflammation.

The cytoplasmic distribution of Weibel-Palade bodies and tissue-specific organelles in EA.hy926 cells, as observed through electron photomicrographs, reflects their differentiated endothelial cell functions. One of the critical advantages of EA.hy926 cells is their ability to undergo more than 100 population doublings (PDLs) while maintaining their cellular properties.

This longevity ensures a sustainable and consistent cell source for long-term experiments and investigations. With a doubling time of 12 hours, these cells exhibit rapid proliferation, facilitating experimental workflows and enabling efficient generation of cell quantities required for large-scale studies.

EA.hy926 cells have proven to be a game-changer in cardiovascular research, particularly in the purification of endothelin converting enzyme (ECE). Traditionally, obtaining primary endothelial cells in significant quantities has been challenging, hindering the sanctification of ECE.

However, EA.hy926 cells, derived from transformed human umbilical vein endothelial cells, have emerged as a reliable alternative for studying ECE activity. This breakthrough has opened up new possibilities for investigating the roles of ECE in cardiovascular diseases and developing potential therapeutic interventions.

**Organism** Human

**Tissue** Umbilical vein, vascular endothelium

**Synonyms** EA. hy 926, EA hy 926, EA-hy926, EAhy 926, EAHY-926, EA.Hy926, EA.hy926, EAhy926, EaHy926, Eahy926

**Gender** Male

**Morphology** Endothelial

**Growth properties** Adherent

**Citation** EA.hy926 (Cytion catalog number 305034)

**Biosafety level** 1

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<b>NCBI_TaxID</b>	9606
<b>CellosaurusAccession</b>	CVCL_3901
<b>Culture Medium</b>	DMEM, w: 4.5 g/L Glucose, w: 4 mM L-Glutamine, w: 3.7 g/L NaHCO <sub>3</sub> , w: 1.0 mM Sodium pyruvate (Cytion article number 820300a)
<b>Supplements</b>	Supplement the medium with 10% FBS
<b>Dissociation Reagent</b>	Accutase
<b>Doubling time</b>	12 hours
<b>Subculturing</b>	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.
<b>Fluid renewal</b>	2 to 3 times per week
<b>Freeze medium</b>	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°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<sub>2</sub>, 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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**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.