



General information

Description Established from the malignant ascites (with peritoneal metastasis).

Organism Human

Tissue Bone

Disease Ewing's Sarcoma

Metastatic site Ascites

Synonyms MHH-ES-1, MHHES1

Characteristics

Age 12 years

Gender Male

Ethnicity Turkish

Morphology Small round cells

Growth properties

Adherent, clusters

Identifiers / Biosafety / Citation

Citation MHH-ES1 (Cytion catalog number 300136)

Biosafety level 1

Depositor Hartmann

Expression / Mutation

Handling



MHH-ES1 Cells | 300136

| Culture Medium | RPMI 1640, w: 2.1 mM stable Glutamine, w: 2.0 g/L NaHCO3 (Cytion article number 820700a) |
|-----------------------|---|
| Medium supplements | Supplement the medium with 10% FBS |
| Passaging solution | 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. |
| Split ratio | A ratio of 1:3 is recommended |
| Seeding density | 1 to 2 x 10^4 cells/cm^2 |
| Fluid renewal | Every 3 to 5 days |
| Freezing recovery | After thawing, plate the cells at 5×10^4 cells/cm 2 and allow the cells to recover from the freezing process and to adhere for at least 24 hours. |
| Freeze medium | CM-1 (Cytion catalog number 800100) or CM-ACF (Cytion catalog number 806100) |



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Handling of cryopreserved cultures

- 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. Optionally, skip centrifugation but remove any remaining freezing medium after 24 hours.
- 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.

Quality control / Genetic profile / HLA

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.



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STR profile CSF1PO: 11

D16S539: 11 D5S818: 13 D7S820: 9,11 TH01: 8,9 TPOX: 8 vWA: 16,17 D3S1358: 15,16 D21S11: 28,32.2 D18S51: 14,16 Penta E: 11,15 Penta D: 11,12 D8S1179: 11,13 FGA: 22

D13S317: 8

HLA alleles A*: 01:01:01, 68:01:01

C*: 01:02:01, 07:01:01

DRB1*: 07:01:01, 11:01:01

DQA1*: 02:01:01, 05:05:01

DQB1*: 03:01:01, 03:03:02G

DPB1*: 10:01:01, 13:01:01

E: 01:01:01, 01:03:01

B*: 40:01:02, 49:01:01