

HS-695T Cells | 300211**General information**

Description HS-695T was isolated in 1973 from the lymph node metastasis of an amelanotic melanoma by A. Creasey et al. The cells had a doubling time of approximately 48 hours at passages 19 through 40 and grew moderately in semi-solid medium.

Organism Human

Tissue Skin

Disease Amelanotic melanoma

Metastatic site Lymph node

Synonyms Hs 695.T, Hs-695-T, Hs 695T, HS 695T, Hs695T, HS695T, Hs695

Characteristics

Age 26 years

Gender Male

Ethnicity Caucasian

Morphology Epithelial-like

Growth properties Adherent

Identifiers / Biosafety / Citation

Citation HS-695T (Cytion catalog number 300211)

Biosafety level 1

Depositor R. B. Owens

Expression / Mutation

Protein expression p53 positive

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Isoenzymes G6PD, B, PGM1, 1, PGM3, 1, ES-D, 1, Me-2, 0, AK-1, 1, GLO-1, 1, Phenotype Frequency Product: 0.0427

Tumorigenic Yes, in immunosuppressed mice

Mutational profile BRAF V600Emut

Karyotype (P19-40) mode = 52, Y chromosome present

Handling

Culture Medium DMEM, w: 4.5 g/L Glucose, w: 4 mM L-Glutamine, w: 1.5 g/L NaHCO₃, w: 1.0 mM Sodium pyruvate (Cytion article number 820300a)

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 1:2 to 1:4 is recommended

Seeding density 2×10^4 cells/cm²

Fluid renewal 2 to 3 times per week

Freezing recovery After thawing, plate the cells at 5×10^4 cells/cm² 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

Amelogenin: x,y
CSF1PO: 11
D13S317: 12
D16S539: 9,13
D5S818: 9
D7S820: 9,10
TH01: 6
TPOX: 8
vWA: 18
D3S1358: 15
D21S11: 29
D18S51: 18
Penta E: 5,11
Penta D: 9,12
D8S1179: 13,15
FGA: 21,24