

HEp-2 Hs 578T | 305089

Thawing and Culturing Cells

1. **Thawing:** Thaw the vial rapidly in a water bath at 37°C. Transfer the cells to a pre-warmed T75 flask containing 15 ml of complete medium. Centrifuge at 300 x g for 3 minutes. Remove the supernatant and resuspend the cells in 10 ml of complete medium. Seed the cells into a T25 flask containing 10 ml of complete medium. Incubate at 37°C with 5% CO₂ until the cells reach 70% confluency.
2. **Passaging:** When the cells reach 70-80% confluency, they can be passaged. Remove the medium and wash the cells with PBS. Add 3 ml of trypsin-EDTA solution and incubate at 37°C for 5 minutes. Add 15 ml of complete medium to stop the trypsin. Detach the cells by scraping the flask with a rubber policeman. Centrifuge at 300 x g for 3 minutes. Resuspend the cells in 10 ml of complete medium. Seed the cells into a T25 flask containing 10 ml of complete medium. Incubate at 37°C with 5% CO₂ until the cells reach 70% confluency.
3. **Freezing:** When the cells reach 70-80% confluency, they can be frozen. Remove the medium and wash the cells with PBS. Add 3 ml of trypsin-EDTA solution and incubate at 37°C for 5 minutes. Add 15 ml of complete medium to stop the trypsin. Detach the cells by scraping the flask with a rubber policeman. Centrifuge at 300 x g for 3 minutes. Resuspend the cells in 1 ml of freezing medium. Seed the cells into a 1.5 ml microcentrifuge tube. Freeze the cells at -80°C.
4. **Storage:** Store the cells at -80°C for long-term storage.
5. **Thawing:** Thaw the vial rapidly in a water bath at 37°C. Transfer the cells to a pre-warmed T75 flask containing 15 ml of complete medium. Centrifuge at 300 x g for 3 minutes. Remove the supernatant and resuspend the cells in 10 ml of complete medium. Seed the cells into a T25 flask containing 10 ml of complete medium. Incubate at 37°C with 5% CO₂ until the cells reach 70% confluency.
6. **Passaging:** When the cells reach 70-80% confluency, they can be passaged. Remove the medium and wash the cells with PBS. Add 3 ml of trypsin-EDTA solution and incubate at 37°C for 5 minutes. Add 15 ml of complete medium to stop the trypsin. Detach the cells by scraping the flask with a rubber policeman. Centrifuge at 300 x g for 3 minutes. Resuspend the cells in 10 ml of complete medium. Seed the cells into a T25 flask containing 10 ml of complete medium. Incubate at 37°C with 5% CO₂ until the cells reach 70% confluency.
7. **Freezing:** When the cells reach 70-80% confluency, they can be frozen. Remove the medium and wash the cells with PBS. Add 3 ml of trypsin-EDTA solution and incubate at 37°C for 5 minutes. Add 15 ml of complete medium to stop the trypsin. Detach the cells by scraping the flask with a rubber policeman. Centrifuge at 300 x g for 3 minutes. Resuspend the cells in 1 ml of freezing medium. Seed the cells into a 1.5 ml microcentrifuge tube. Freeze the cells at -80°C.
8. **Storage:** Store the cells at -80°C for long-term storage.

Incubation Atmosphere

37°C, 5% CO₂, humidified

Flask Coating

Flasks are pre-coated with poly-L-lysine.

Freezing Procedure

When the cells reach 70-80% confluency, they can be frozen. Remove the medium and wash the cells with PBS. Add 3 ml of trypsin-EDTA solution and incubate at 37°C for 5 minutes. Add 15 ml of complete medium to stop the trypsin. Detach the cells by scraping the flask with a rubber policeman. Centrifuge at 300 x g for 3 minutes. Resuspend the cells in 1 ml of freezing medium. Seed the cells into a 1.5 ml microcentrifuge tube. Freeze the cells at -80°C.

Shipping Conditions

Shipping conditions: -78°C

Storage Conditions

Storage conditions: -150 to 196 K

Genotype / Phenotype / HLA

Sterility

HEp-2 cells are tested for mycoplasma contamination using PCR.

HEp-2 cells are tested for mycoplasma contamination using PCR.