

HaCaT | 300493

Thawing and Culturing Cells

1. Thaw the vial in a water bath at 37°C. Centrifuge at 300 x g for 3 minutes. Remove the supernatant and resuspend the cells in 10 ml of DMEM supplemented with 10% FBS. Seed the cells into a T75 flask.
2. Incubate the cells until they reach 70-80% confluency. Then, replace the medium with DMEM supplemented with 1% FBS for 24 hours to allow the cells to reach a steady state.
3. Harvest the cells by trypsinization and seed them into a new T75 flask with fresh DMEM supplemented with 10% FBS.
4. Repeat the process until you have a sufficient number of cells for your experiment.
5. For long-term storage, harvest the cells and seed them into a T75 flask with DMEM supplemented with 10% FBS. Once they reach 70-80% confluency, harvest them and resuspend them in 1 ml of DMEM supplemented with 10% FBS. Seed them into a 1.5 ml microcentrifuge tube and centrifuge at 300 x g for 3 minutes. Remove the supernatant and resuspend the cells in 100 µl of DMEM supplemented with 10% FBS. Store the cells at -150°C.
6. For long-term storage, harvest the cells and seed them into a T75 flask with DMEM supplemented with 10% FBS. Once they reach 70-80% confluency, harvest them and resuspend them in 1 ml of DMEM supplemented with 10% FBS. Seed them into a 1.5 ml microcentrifuge tube and centrifuge at 300 x g for 3 minutes. Remove the supernatant and resuspend the cells in 100 µl of DMEM supplemented with 10% FBS. Store the cells at -150°C.
7. For long-term storage, harvest the cells and seed them into a T75 flask with DMEM supplemented with 10% FBS. Once they reach 70-80% confluency, harvest them and resuspend them in 1 ml of DMEM supplemented with 10% FBS. Seed them into a 1.5 ml microcentrifuge tube and centrifuge at 300 x g for 3 minutes. Remove the supernatant and resuspend the cells in 100 µl of DMEM supplemented with 10% FBS. Store the cells at -150°C.
8. For long-term storage, harvest the cells and seed them into a T75 flask with DMEM supplemented with 10% FBS. Once they reach 70-80% confluency, harvest them and resuspend them in 1 ml of DMEM supplemented with 10% FBS. Seed them into a 1.5 ml microcentrifuge tube and centrifuge at 300 x g for 3 minutes. Remove the supernatant and resuspend the cells in 100 µl of DMEM supplemented with 10% FBS. Store the cells at -150°C.

Incubation Atmosphere

37°C, 5% CO₂, humidified

Flask Coating

None

Freezing Procedure

Harvest the cells and seed them into a T75 flask with DMEM supplemented with 10% FBS. Once they reach 70-80% confluency, harvest them and resuspend them in 1 ml of DMEM supplemented with 10% FBS. Seed them into a 1.5 ml microcentrifuge tube and centrifuge at 300 x g for 3 minutes. Remove the supernatant and resuspend the cells in 100 µl of DMEM supplemented with 10% FBS. Store the cells at -150°C.

Shipping Conditions

Store the cells at -150°C. Ship the cells on dry ice.

Storage Conditions

Store the cells at -150°C. Ship the cells on dry ice.

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Sterility

The cells are provided as a frozen stock. The cells are tested for mycoplasma contamination using PCR. The cells are free of mycoplasma contamination.

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HLA **A***: '31:01:02
B*: '40:01:02, '51:01:01
C*: '03:04:01, '15:02:01
DRB1*: '04:01:01, '15:01:01
DQA1*: '01:02:01, '03:03:01
DQB1*: '03:01:01, '06:02:01
DPB1*: '03:01:01, '04:01:01
E: '01:03:01, '01:03:02