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User's Guide

Human Umbilical Cord Blood Mesenchymal Stem Cells

The following describes the preferred methods for the thawing and subsequent handling and culturing of human umbilical cord blood mesenchymal stem cells.

Thaw Procedure
NOTE: Cells should be kept on dry ice until ready for use. If necessary, transfer frozen cells for further storage into a -85°C freezer. It is not recommended to place cells into liquid nitrogen (LN_2) storage.

Cells are provided in vials within a protective overwrap to ensure purity. This overwrap also causes some thermal lag during the thaw procedure. For these reasons, vials should be thawed in a shallow 40°C water bath. Place the vial into the bath and allow it to warm until the last bit of ice just melts.

Spray the outer container with ethanol and dry. Using scissors, cut away the overwrap and place the vial into the hood. Remove the cap and using a pipette, transfer the contents of the vial into a 15mL conical tube.

Add 100µL/minute of culture medium to the thawed sample each minute for 10 minutes (the final volume should be around 2 mL).

After the 10 additions, wait one minute and add an additional 5mL of culture medium (the final volume should now be around 7mL).

Wait 5 minutes and then centrifuge the cells at 500 x g for 5 minutes.

Aspirate the supernatant, resuspend the cells for culture.

Culture Procedure
Seed cells at a density of 5000-6000 cells/cm^2 on tissue culture treated plates or flasks

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It is recommended to feed cells every 3-4 days, and cells typically are ready for subculture (passage) by day 7.

It is recommended that cells be passaged at not more than 70% confluence to maintain an undifferentiated state.

Quality Control
Each cell line is performance assayed post thaw. A certificate of analysis (CoA) for each line is shipped with each order. Non-routine performance and quality testing to meet your specifications is available for an additional fee.
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