CD34+ Progenitor Cells

PromoCell®

Instruction Manual

Product	Size	Catalog Number
Human CD34+ Progenitor Cells from Cord Blood (hCD34+-CB) single donor	100,000 cryopreserved cells	C-12921

Product Description

CD34 is a glycosylated transmembrane protein and represents a well-known marker for primitive blood- and bone marrow-derived progenitor cells, especially for hematopoietic and endothelial progenitors. Although the biological functions of CD34 are largely unknown, recent data suggest that CD34 is involved in maintenance of the progenitor cells in a phenotypically undifferentiated state. PromoCell offers CD34+ Progenitor Cells from the cord blood of healthy donors, produced at PromoCell's cell culture facility. CD34+ Progenitor Cells are suitable for a series of studies, e.g. directed differentiation into more committed types of blood cells and endothelial lineages.

Immediately after isolation, the freshly prepared CD34+ Progenitor Cells are cryopreserved using PromoCell´s proprietary serum-free freezing medium Cryo-SFM. Each cryo vial contains more than 100,000 viable cells after thawing.

Quality Control

Rigid quality control tests are performed for each lot of PromoCell CD34+ Progenitor Cells.

They are routinely characterized by flow cytometry analyzing the viability, cell size, and the presence of significant markers, i.e. CD34.

In addition, all cells have been tested for the absence of HIV-1, HIV-2, HBV, HCV, HTLV-1, HTLV-2 and microbial contaminants (fungi, bacteria, and mycoplasma). A detailed certificate of analysis (CoA) for each lot can be downloaded at: www.promocell.com/coa

Intended Use

PromoCell CD34+ Progenitor Cells are for *in vitro* research use only and not for diagnostic or therapeutic procedures.

Warning

Although tested negative for HIV-1, HIV-2, HBV, HCV, HTLV-1 and HTLV-2, the cells – like all products of human origin – should be handled as potentially infectious. No test procedure can completely guarantee the absence of infectious agents.

Follow appropriate safety precautions!

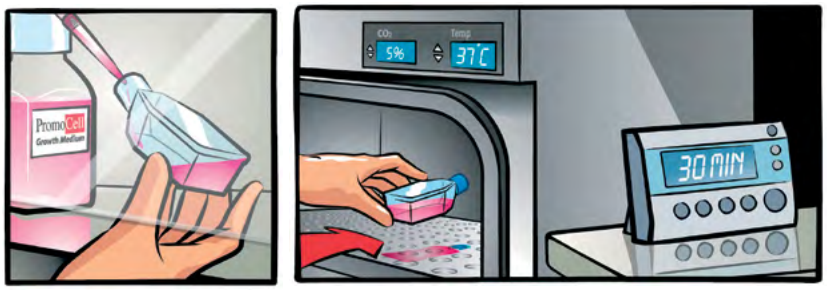
After delivery, start immediately with the protocol for cryopreserved cells (see page 2).

Start immediately after delivery.
Use aseptic techniques and a laminar flow bench.

Protocol for Cryopreserved Cells

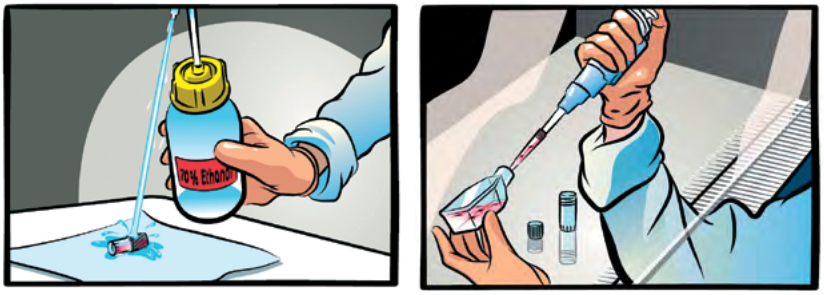
Straight after arrival, store the cryopreserved cells in liquid nitrogen or seed them immediately.
Note: Storage at -80°C is not sufficient for cell preservation and causes irreversible cell damage.

1. **Prepare the medium**
   Transfer the needed volume of PromoCell Medium according to the seeding density (see page 4) in cell culture vessels. Use at least 9 ml medium per vial of cells. For equilibration, place the vessels in an incubator (37°C, 5% CO_2) for 30 minutes.



2. **Thaw the cells**
   Remove the cryovial from the liquid nitrogen container and immediately place it on dry ice - even for short transportation. Under a laminar flow bench, briefly twist the cap a quarter turn to relieve pressure, then retighten. Immerse the vial into a water bath (37°C) just up to the screw cap for 2 minutes. Ensure that no water enters the thread of the screw cap.

3. **Disinfect the vial and seed the cells**
   Thoroughly rinse the cryovial with 70% ethanol under a laminar flow bench. Then, aspirate the excess ethanol from the thread area of the screw cap. Open the vial and transfer the cells to a cell culture vessel containing the prewarmed medium from step 1.

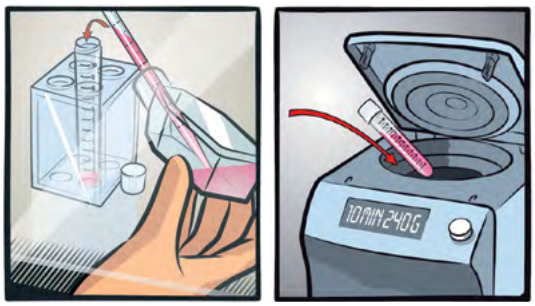


4. **Incubate the cells**
   Place the vessel in an incubator (37°C, 5% CO2). Replace the medium after 4–8 hours according to the subcultivation protocol (see page 3). The cells should be subcultured as required.

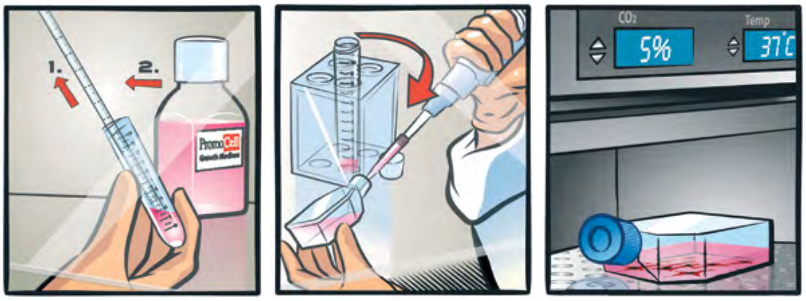
Use aseptic techniques and a laminar flow bench.

Subcultivation Protocol

1. **Harvest the cells**
   Harvest the cell suspension and determine the cell number. Spin down the cells for 10 minutes at 240 x g.



2. **Resuspend and reseed cells**
   Discard the supernatant (step 1), add 1 ml of the appropriate PromoCell Medium (step 2), and resuspend the cells by carefully pipetting up and down. Seed the cells according to the recommended seeding density in new cell culture vessels containing fresh PromoCell Medium prewarmed to 37°C. Place the vessels in an incubator (37°C, 5% CO_2).



Specifications

Product	Recommended Culture Media	Plating density
Human CD34⁺ Progenitor Cells from Cord Blood (hCD34⁺-CB), single donor	C-28021 C-39891	20,000 cells per ml

Related Products

Product	Size	Catalog Number
Hematopoietic Progenitor Cell Expansion Medium DXF	500 ml	C-28021
Cytokine Mix E for HPC Expansion Medium DXF	1 ml (sufficient for 100 ml Medium) 5 ml (sufficient for 500 ml Medium)	C-39890 C-39891
Cryo-SFM	30 ml 125 ml	C-29910 C-29912
G-CSF, human, recombinant	10 µg	C-60434
SCF, human, recombinant	10 µg	C-63120
flt3-ligand, human, recombinant	10 µg	C-67110
IL-3, human, recombinant	30 µg	C-61320
IL-6, human, recombinant	10 µg	C-61625
TPO, human, recombinant	10 µg	C-65112

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