



iPSC-Derived Human Hematopoietic Progenitor Cell (HPC) Kit (African-American, Male Line)

Product Information

Catalog Number ASE-9718

Description Applied StemCell has developed an efficient integration-free, small molecule-based method to differentiate high-quality hematopoietic progenitor cells (HPC) from human iPSCs. The differentiated HPCs recapitulate the phenotype and functional parameters of primary and *in vivo* HPCs.

We provide HPCs differentiated from an integration-free, control human iPSC line (ASE-9211), reprogrammed from the fibroblasts of an African-American male donor. These high-purity ($\geq 70\%$) cells express high levels of HPC biomarker, CD34 (Figure 1).

To harness the full potential of our HPCs, we also provide optimized HPC Culture Basal Media (ASE-9718MM) that supports robust maintenance and functionality of the HPCs in culture.

These iPSC-differentiated HPCs can be used as control lines to compare phenotype and functionality of patient-derived, genome edited iPSC-derived HPCs for drug screening applications.

Parental Tissue Control human iPSC (ASE-9211); p15
Age: Neonate
Gender: Male
Ethnicity: African-American
Tissue Source: Dermal Fibroblasts
Reprogramming Method: Episomal
Culture Conditions: Feeder-free

Clinical information Healthy (with no known disease phenotypes)

Shipping Dry ice

Storage and Stability Store the components of the kit at the appropriate storage conditions as indicated in the media and materials table, immediately upon arrival. Shelf-life of the product is contingent upon proper storage conditions

Quality Control Each lot of iPSC-derived human HPCs has been tested for growth, viability and purity ($\geq 70\%$) following recovery from cryopreservation. In addition, each lot has been tested for expression of HPC markers and for the absence of mycoplasma and pathogens.

Safety Precaution **PLEASE READ BEFORE HANDLING ANY FROZEN VIALS.** Please wear appropriate Personal Protection Equipment (lab coat, thermal gloves, safety goggles and a face shield) when handling frozen vials. Please be aware that the following scenario can occur: Liquid nitrogen can leak into the vials when the vials are submerged in liquid nitrogen. Upon thawing, the liquid nitrogen returns to the gas phase, resulting in a dangerous build-up of pressure within the vial. This can result in

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the vial exploding and expelling not only the vial contents but also the vial cap and plastic fragments of the vial.

Warranty

The performance of Applied StemCell's iPSC-derived HPCs has been validated with the HPC Culture Basal Media provided in the Hematopoietic Progenitor Cell Kit and the recommended additional reagents. Applied StemCell will not hold responsibility if components other than the components provided with the Hematopoietic Progenitor Cell Kit and those recommended are used to culture the Applied StemCell HPCs.

Restricted Use

This product is for research use only and not intended for human or animal diagnostic or therapeutic uses.

Characterization of the ASE-9718 Hematopoietic Progenitor Cells

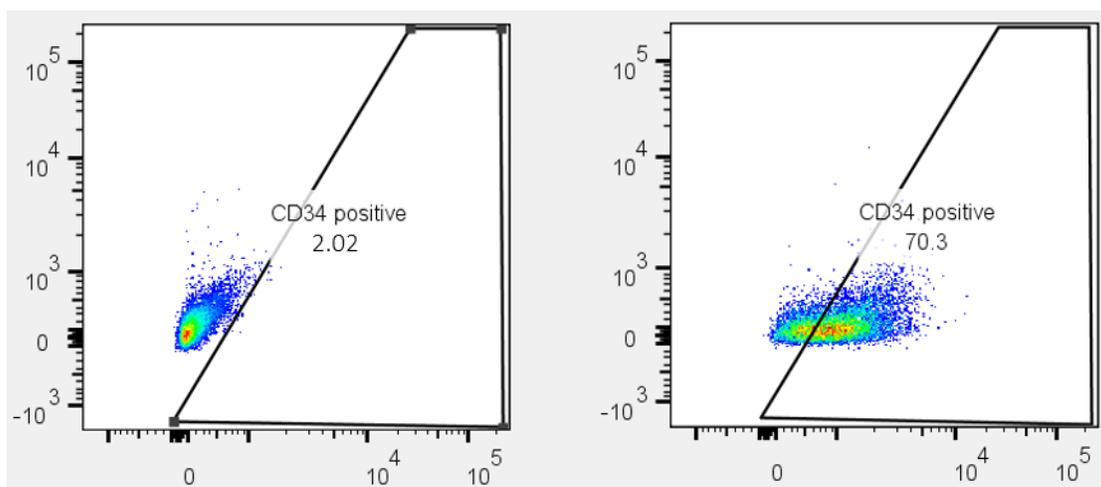


Figure 1. Flow cytometry analysis of ASE-9718 iPSC-derived Hematopoietic Progenitor Cells for an HPC biomarker. Cryopreserved HPCs, differentiated from Applied StemCell's control iPSC line, ASE-9211 were recovered in HPC culture basal media. The cells were stained with HPC cell marker, CD34 (right panel) or isotype control (Left panel).

Media and Material

Hematopoietic Progenitor Cell Kit (ASE-9718)

Catalog #	Component	Amount	Storage	Shelf Life
ASE-9718-C	iPSC-derived Hematopoietic Progenitor Cells; African-American Male Line	≥1x10 ⁶ cells/ vial	Liq. N2	12 months
ASE-9718MM	HPC Culture Basal Media	100 mL	-20°C	12 months

Additional Reagents Required

The below reagents are recommended for use with the HPCs. If you use reagents other than those recommended, we suggest that you do a batch-test to validate quality of the cells and culture protocol.

- Primary antibodies:
 - CD34 Biolegend #343603

Protocol

1. Preparing Cell Culture Surfaces

- 1.1 Please use ultra-low attachment vessels for best results.

2. Preparation of Hematopoietic Progenitor Cell Culture Media

- 2.1 Thaw the HPC Culture Basal Media at room temperature before thawing the cryopreserved HPCs.
- 2.2 Add required hematopoietic cytokines and growth factors into HPC Culture Basal Media for intended use.

3. Thawing and Culturing Cryopreserved Hematopoietic Progenitor Cells

- 3.1 To thaw the cryopreserved HPCs, remove one vial from the storage unit.
- 3.2 Immerse the vial in the water bath (up to 2/3rd of the vial) and thaw the cells rapidly until only a small piece of ice is still visible (approximately one minute).
Note: Do not shake the vial during thawing.
- 3.3 Bring the vial to the biological cabinet immediately and spray the outside of the vial thoroughly with 70% ethanol and wipe it with an autoclaved paper towel.
- 3.4 Remove the cells from the vial using a p1000 micropipette (or serological pipette) and transfer it slowly, drop-wise while swirling into a 15 mL conical tube containing 5 mL of pre-warmed HPC Culture Medium. Wash the vial with 1 mL medium from the 15 mL conical tube and transfer it back to the tube.
Note: Do not mix cells up and down and avoid generation of bubbles.
- 3.5 Centrifuge cells at 250 x g for 5 minutes at room temperature.
- 3.6 Aspirate the medium very carefully using a vacuum (or pipette if preferred), leaving only a drop of liquid in the tube.
Note: Take extra care not to remove or disturb the cell pellet during aspiration of medium.
- 3.7 Using a p1000 micropipette, add 1 mL of the pre-warmed HPC Culture Medium into the tube and gently re-suspend cells by pipetting up and down 2-3 times.
- 3.8 Remove a 10 μ L aliquot of the cell suspension and mix it with 10 μ L of Trypan blue solution.
- 3.9 Count the cells.
- 3.10 Aspirate the coating matrix from the pre-warmed cell culture vessel.
- 3.11 Seed the HPCs at desired plating density.
- 3.12 Distribute the cells evenly.
- 3.13 Place the cell culture vessels in the incubator (37°C/ 5% CO₂/ humidity control).