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Data Sheet

Normal Human Peripheral Blood Mononuclear Cells, Frozen

Catalog # 79059

Description

Cryopreserved vial (30 x 10⁶ cells) of freshly isolated primary human peripheral blood mononuclear cells (PBMCs) from a healthy donor, isolated from whole blood or leukapheresis samples using Ficoll gradient. After isolation, the PBMCs were stained to identify sub populations and evaluated for viability by flow cytometry. Cells were cryopreserved in serum-free Cryostor CS10 at a controlled rate.

Source

291mL leukopak blood

Stability and Storage

Store cells at -135°C or colder. Thawed cells should be used immediately for downstream applications. As these are primary cells, we do not recommend maintaining these cells in culture for long periods of time.

Characterization Criteria

Cell count, viability (trypan blue exclusion and FACS with impermeable DNA binding dye), and surface markers (CD45+, CD14+, CD19+, CD3+, CD4+, CD8+, CD56+)

Medium

Thaw Medium 10 (BPS Bioscience #79704): RPMI 1640 medium (Thermo Fisher, #A1049101) supplemented with 10% FBS (Thermo Fisher, #26140079), 1mM Sodium pyruvate (Corning, #25-000-CL), 1% Non-essential amino acids (Corning, #25-025-CL), 1% Penicillin/streptomycin (Thermo Fisher, #15140122)

Materials or Instruments Required but not Supplied:

Red Blood Cell Lysis buffer (if desired): If cell pellets appear pinkish/light red it is due to a small fraction of Red Blood Cells that did not isolate from the PBMC layer. If desired/necessary, these can be quickly and easily lysed while leaving the PBMC intact, by using RBC Lysis Buffer. RBC may not interfere with downstream PMBC culture experiments, but this will depend on the assay.

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Handling Directions

It is important to work quickly in the following steps to ensure high cell viability and recovery. Quickly thaw cells in a 37°C water bath with constant and slow agitation until >90% thaw. Clean the outside of the vial with 70% ethanol, then immediately transfer the entire contents of the tube into a 15mL vial, rinsing once with medium (Thaw Medium 10). Slowly add 10 ml of pre-warmed medium to the 15mL vial. Gently mix the tube by inverting and centrifuge the cell suspension at 300 x g for 10 minutes at room temperature. Carefully remove the supernatant with a pipette without disturbing the pellet. Wash the cells once more by gently resuspending the cell pellet in 10mL warm medium. Centrifuge the cell suspension at 300 x g for 10 minutes at room temperature. Carefully remove the supernatant with a pipette without disturbing the pellet. NOTE: Up to 20% of cell loss can be expected during washing steps. Cells are now ready for use in downstream applications.

Donor Screening

Donors have been screened and determined negative for:

- Hepatitis B (anti-HBc EIA, HBsAg EIA)
- Hepatitis C (anti-HCV EIA)
- Human Immunodeficiency Virus (HIV-1/HIV-2 plus O)
- Human T-Lymphotropic Virus (HTLV-I/II)
- HIV-1/HCV/HBV
- West Nile Virus
- Trypanasoma cruzi

Note: Testing cannot completely guarantee that any sample is completely virus-free. These cells should be treated as potentially infectious and appropriate biological safety level 2 precautions should be used.

Donor Demographics and Lot Specific Information

Donor Gender	Age	Ethnicity	Blood Type	Cryopreservation Date
Male	29	African American	O-POS	Oct 1, 2019

Surface Marker Summary

7-AAD Viability Dye	CD14 ⁺ Monocytes	CD19 ⁺ B cells	CD3 ⁺ T Cells	CD4 ⁺ T Cells*	CD8 ⁺ T Cells*	CD45 ⁺ Cells	CD56 ⁺ NK Cells
94.0% (live)	14.4%	19.1%	44.3%	35.4%	45.4%	84.1%	2.6%

*CD4 and CD8 percentages are of total CD3+ gated T cells

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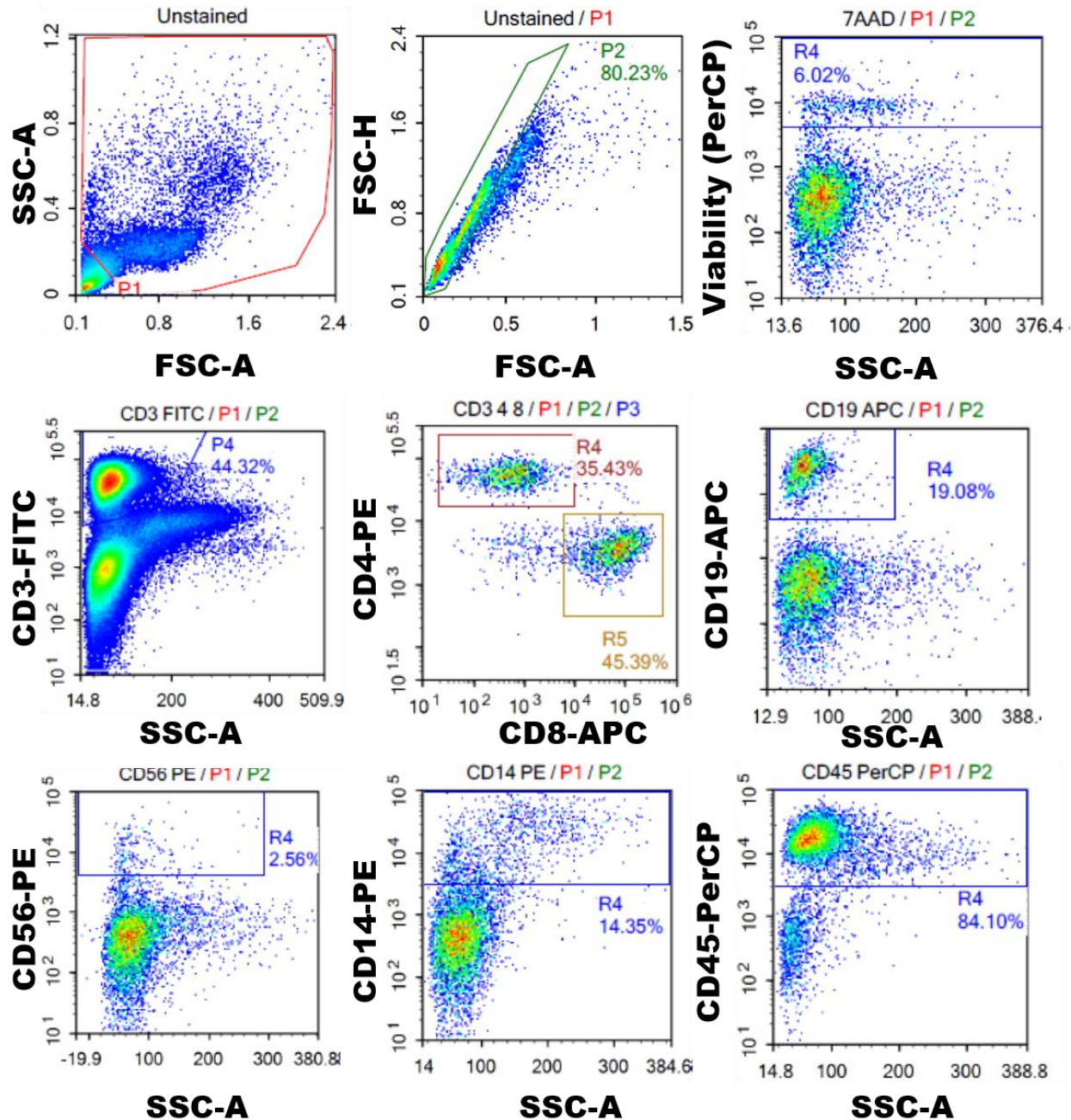
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Flow Cytometry Analysis

Results from frozen cells that were thawed and washed according to the handling directions



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