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Multiparametricimmunophenotyping of human hematopoietic stem cells and progenitor cells by flow cytometry

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Umbilical cord blood, peripheral blood, and bone marrow are the major sources of hematopoietic stem cells (HSCs) and progenitor cells. The characterization and enumeration of HSCs and progenitor cells from these samples can provide valuable information for clinical research. Multiparametric flow cytometry is a well-established method for immunophenotyping of HSCs and various subpopulations of the progenitor cells. Using 8-color panels, a lyse/no-wash assay was developed to analyze cell surface phenotypic markers of HSCs and the various progenitor cell populations, such as multipotent progenitor cells (MPPs), common myeloid progenitor cells (CMPs), common lymphoid progenitor cells (CLPs), megakaryocyte erythroid progenitor cells (MEPs), and granulocyte macrophage progenitor cells (GMPs). In addition, regulatory T cells (Tregs) and mesenchymal stem cells (MSCs) were also measured. These cell populations are reported to be functionally important in a transplant blood unit, and may influence engraftment success and the development of graft-versus-host disease in the recipient.

During the development of this research assay, cord blood and bone marrow samples were tested foridentification and enumeration of cells in each of the subpopulations. Additionally, enumeration of $CD34^+$ cells was also compared with the 3-color $CD34^+$BD™ Stem Cell Enumeration (SCE) assay using a limited sample set. The percent $CD34^+$ data from the samples tested showed less than 10% difference between the 8-color and 3-color assays. Using this assay, we tested 30 cryopreserved attached segments from cord blood units and investigated the aforementioned cell populations. The frequency of each cell population in the attached segments reflects the frequency of cells in the cord blood units themselves.

Cell count data revealed variations in the HSC and progenitor subpopulations from sample to sample, which might have a significant impact on the recovery rate of neutrophils, platelets, and the overall immune system in stem cell recipients after transplantation. Overall, these 8-color panels could be used as a valuable research tool for characterization and enumeration of HSCs and progenitor cells in research transplantation units.

Biography

Phil Paul serves as Executive Officer and Director of the Cleveland Cord Blood Center and has been a member of the management team since the Center's inception in May of 2007. He received his Ph.D. in Experimental Hematology from New York University, and completed post doctoral training and a fellowship in Laboratory Hematology at the Cleveland Clinic; followed by 25 years on the professional staff in the Departments of Laboratory Medicine, the Research Institute, and the HLA Laboratory. He is board certified (ABB) in Molecular Diagnostics.

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