Solutions for cutting-edge human hematopoietic stem cell research
Isolate hematopoietic stem cells (HSCs) from various starting materials with MACS® Technology and achieve highly pure samples by choosing the right kit for your downstream applications.

- CD34 MicroBead Kit UltraPure, human for debris-rich samples (fig. 1).
- CD34+CD38– Cell Isolation Kit, human for primitive HSC enrichment.
- Diamond CD34 Isolation Kit, human combines lineage depletion and CD34+ enrichment.
- CD133 MicroBead Kit Hematopoietic Tissue, human – for CD133 enrichment of primitive and early HSCs.

**Table 1:** Indicated for isolation out of ⚫ umbilical cord blood, bone marrow, apheresis product and ⚫ peripheral blood, and differentiated ES and iPS cells.


- Xeno- and serum-free basic medium.
- Maintains high levels of CD34+ cells.
- Preserves characteristic surface phenotype of early HSCs.

**Table 2:**

<table>
<thead>
<tr>
<th>Products</th>
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<tbody>
<tr>
<td>CD34 MicroBead Kit, human</td>
<td>130-046-702</td>
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<tr>
<td>CD34 MicroBead Kit UltraPure, human</td>
<td>130-100-453</td>
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<tr>
<td>CD34+CD38– Cell Isolation Kit, human</td>
<td>130-114-822</td>
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<tr>
<td>Diamond CD34 Isolation Kit, human</td>
<td>130-094-531</td>
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<tr>
<td>CD133 MicroBead Kit – Hematopoietic Tissue, human</td>
<td>130-100-830</td>
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**Figure 1:** Isolation of CD34+ cells from a PBMC sample using the CD34 MicroBead Kit UltraPure. Flow cytometric analysis shows high purity after enrichment.

**Figure 2:** CD34+ cord blood cells were expanded for seven days in StemMACS HSC Expansion Media XF supplemented with StemMACS HSC Expansion Cocktail. The cell surface phenotype was assessed by flow cytometry.
The transduction enhancer Vectofusin-1® streamlines the transduction of HSCs with retroviral vectors (fig. 3).

- Water-soluble synthetic peptide.
- Unlike recombinant fibronectin, pre-coating is not required.
- High cell viability.

Analyze HSCs with our constantly growing portfolio of antibodies for the detection of HSC markers, such as CD34, CD38, CD133, and CD117.

- REAfinity™ Recombinant Antibodies make Fcγ receptor blocking obsolete requiring only one isotope control, saving time and effort.
- VioDye® Family of fluorochromes for multicolor flow cytometric analysis with high fluorescence intensities and low spillover.
- REAlease® Fluorochrome Technology for removal of any labels after cell sorting.

**Figure 3:** Magnetically isolated CD34+ cells were transduced with lentiviral vectors (LVs) encoding GFP at a multiplicity of infection (MOI) of 10 without facilitating agent, in the presence of Vectofusin-1 or in recombinant fibronectin–coated plates. LVs displayed baboon envelope (BaEV) or measles virus (MV) pseudo types. Two different BaEV constructs were used: BaEV 1 and BaEV 2. Vectofusin-1, which does not require pre-coating, enhances the transduction similar to recombinant fibronectin.

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<tr>
<td>Vectofusin-1</td>
<td>130-111-163</td>
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Colony-forming unit (CFU) assays help to examine the ability of HSCs to proliferate and differentiate into colonies in response to cytokine stimulation. Analysis can be performed by visual scoring with the help of a microscope or by flow cytometric analysis.

- StemMACS™ HSC-CFU Media are semi-solid media and can be used for standard CFU assays using visual scoring with a microscope.
- The StemMACS HSC-CFU Assay Kit, human combines differentiation in methylcellulose-free cell culture with a flow cytometric readout for high-throughput and user-independent analysis (fig. 4).

Irrespective of which analysis you choose, the results of our HSC-CFU assays are similar.

**Figure 4:** Workflow of the CFU assay using the StemMACS HSC-CFU Assay Kit, human with flow cytometric analysis. Cells, diluted in StemMACS HSC-CFU Assay Media, are plated in 96-well plates and proliferate and differentiate for 14 days. After the incubation period, cells are stained with the StemMACS HSC-CFU Assay Cocktail and analyzed by flow cytometry. BFU-E: burst-forming unit-erythroid, CFU-G: CFU-granulocyte, CFU-M: CFU-macrophage, CFU-GM: CFU-granulocyte-macrophage, CFU-GEMM: CFU-granulocyte/erythrocyte/macrophage/megakaryocyte.

**Figure 5:** CFU assay results by flow cytometric analysis and visual scoring. CD34+ cells from three different sources (mobilized leukapheresis, cord blood, peripheral blood mononuclear cells [PMBCs]) were seeded in the media from the StemMACS HSC-CFU Assay Kit, human (250 HSCs per 96-well plate) for flow cytometric analysis and the StemMACS HSC-CFU Media (250 HSCs per 35 mm cell culture dish) for visual scoring. Cells were incubated for 14 days and analyzed with the MACSQuant® Analyzer (flow cytometric analysis) or with a microscope (visual scoring). The distribution of the different colony types is highly comparable between both methods.

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<tr>
<td>StemMACS HSC-CFU complete with Epo, human</td>
<td>130-091-280</td>
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<tr>
<td>StemMACS HSC-CFU complete w/o Epo, human</td>
<td>130-091-277</td>
</tr>
<tr>
<td>StemMACS HSC-CFU lite with Epo, human</td>
<td>130-091-281</td>
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<tr>
<td>StemMACS HSC-CFU Assay Kit, human</td>
<td>130-125-042</td>
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Hematopoietic stem cells (HSCs) are tissue-specific adult stem cells capable of differentiating into all blood cell types to ensure homeostasis of blood throughout life. HSCs are used as a model to understand hematopoiesis regulation and to elucidate mechanisms driving stem cell self-renewal or the directed differentiation towards specific blood cell types. Clinically, HSCs or HSC-enriched fractions are used to treat blood related diseases, like leukemia, sickle cell disease, β-thalassemia or the immune defect severe combined immunodeficiency (SCID). HSCs can be isolated from various tissues such as umbilical cord blood, bone marrow, apheresis product, peripheral blood, or differentiated ES and iPS cells. Depending on the starting material, HSC content varies from 0.1 to 2.5%.

HSC workflow – Overview

Hematopoietic stem cell (HSC)

Flowcytometry

Transduction

Expansion

Functionalanalysis

HSC enrichment

Starting material, e.g. blood

Density gradient