

## Cell-Vive™ CD DMSO-Free Cryopreservation Solution, GMP

<b>Catalog# / Size</b>	420516 / 100 mL
<b>Other Names</b>	Cryo Solution, DMSO-free, Cryopreservation media
<b>Description</b>	<p>Cell-Vive™ CD DMSO-Free Cryopreservation Solution, GMP is recommended to support human Mesenchymal Stem Cells (MSCs) and immune cell cryopreservation in a DMSO-free fashion. It is a chemically-defined formulation prepared without human or other animal-derived components. It is suitable for use in the cryopreservation of different MSCs sources, without affecting relevant properties. When used under appropriate conditions, this product can preserve cell viability, expansion, phenotype, and differentiation capacity of MSCs. When used to cryopreserve immune cells, this product preserves cell viability, expansion, phenotype and function. This GMP product is suggested for use in research and <i>ex vivo</i> cell processing use. The benefits of this Cryopreservation solution include:</p> <ul style="list-style-type: none"> <li>• DMSO-free and Chemically-defined formulation.</li> <li>• Effective in cryopreserving Human MSCs from multiple sources.</li> <li>• Able to maintain important cellular properties</li> </ul>

<b>Quality Statement</b>	<p>BioLegend Cell-Vive™ GMP cell culture products are manufactured and tested in accordance with USP Chapter 1043, Ancillary Materials for Cell, Gene and Tissue- Engineered Products and Ph. Eur. Chapter 5.2.12 in a dedicated GMP facility compliant with ISO 13485:2016. Specifications and processes include:</p> <ul style="list-style-type: none"> <li>• Low endotoxin level (&lt;1 EU/mL)</li> <li>• Mycoplasma and bacterial/fungal growth testing</li> <li>• Batch-to-batch consistency</li> <li>• Vendor qualification</li> <li>• Raw material traceability and documentation</li> <li>• Documented procedures and employee training</li> <li>• Equipment maintenance and monitoring records</li> <li>• Lot-specific certificates of analysis</li> <li>• QA review of released products</li> <li>• Quality audits per ISO 13485:2016</li> </ul>
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### Product Details

<b>Formulation</b>	Chemically-defined. Cell-Vive™ CD DMSO-Free Cryopreservation Solution, GMP is a solution ready to use.
<b>Endotoxin Level</b>	< 1 EU/mL
<b>Preparation</b>	DMSO-Free, Chemically-Defined, no preservatives.
<b>Storage &amp; Handling</b>	4°C (2°C - 8°C)
<b>Application</b>	<a href="#">Cryopreservation of Human MSCs - Quality tested</a>
<b>Recommended Usage</b>	Freeze 0.5-5 million cells per 1mL of Cell-Vive™ CD DMSO-Free Cryopreservation Solution, GMP
<b>Application Notes</b>	For cryopreservation of MSCs, a range between 1-5 million cells/mL of Cell-Vive™ CD DMSO-Free Cryopreservation Solution, GMP is recommended. For cryopreservation of immune cells, a range between 1-100 million cells/mL of Cell-Vive™ CD DMSO-Free Cryopreservation Solution, GMP has been demonstrated to have no impact on recovery or post-thaw function.

The appearance of the product in liquid form is clear with a slight pink color.

#### MSCs Cryopreservation Protocol:

1. Prepare an MSC suspension following an appropriate protocol.
2. Centrifuge cells at 300 x g for 5 minutes to obtain a pellet. Carefully aspirate supernatant.
3. Resuspend pelleted cells with appropriate volume of Cell-Vive™ CD DMSO-Free Cryopreservation Solution, GMP and dispense the cell suspension into cryovials.
4. Immediately transfer the cryovials into a cell freezing container (CoolCell® Container or equivalent) and store it at -80°C. Alternatively, follow an equivalent controlled freezing protocol.
5. After 24-36 hours, transfer cryovials into a liquid nitrogen tank for long-term storage.

#### Thawing MSCs Protocol

1. Pre-warm intended cell culture medium or buffer in 37°C
2. Rapidly thaw frozen vials of cells in a 37°C water bath. Remove the vial from the water bath once the majority of the solutions is thawed, the vial should remain cool to touch and a small ice fragment present.

- Wipe external surface of cell container (vials, flask culture) with 70% ethanol and transfer to the biosafety cabinet.
- Add pre-warmed media to the cells and mix gently at 1:1 ratio.
- Count cells to determine total viable cell density using preferred method.
- After counting the viable cells, seed MSCs at 6,000 cells/cm<sup>2</sup> density in desired complete medium in a 37°C, 5% CO<sub>2</sub> incubator.
- Replace medium after 24 hours to remove non-adherent MSCs and debris.

#### Immune Cell Cryopreservation Protocol:

- Prepare a cell suspension at desired concentration following an appropriate protocol.
- Centrifuge cells at 300 x g for 5 minutes to obtain a pellet. Carefully aspirate supernatant.
- Resuspend pelleted cells with appropriate volume of Cell-Vive™ CD DMSO-Free Cryopreservation Solution, GMP and dispense the cell suspension into cryovials.
- Immediately transfer the cryovials into a cell freezing container (CoolCell® Container or equivalent) and store it at -80°C. Alternatively, follow an equivalent controlled freezing protocol.
- After 24 -36 hours, transfer cryovials into a liquid nitrogen tank for long-term storage.

#### Thawing Immune Cells Protocol:

- Pre-warm intended cell culture medium or buffer in 37°C.
- Rapidly thaw frozen vials of cells at 37°C. Remove the vial from the water bath once the majority of the solution is thawed. The vial should remain cool to touch and a small ice fragment present.
- Wipe external surface of cell container (vials, flask culture) with 70% ethanol and transfer to the biosafety cabinet.
- Gently pipette the thawed cell suspension media and add to a desired volume of pre-warmed cell culture medium.
  - Optional: Take 1 mL of cell culture medium and wash the walls of the cryovial to obtain additional cells. While this is not required, it is recommended to ensure full recovery of frozen cells.
- Gently mix the cell suspension to obtain a homogenous mixture. Count and plate cells using desired protocol.

#### Disclaimer

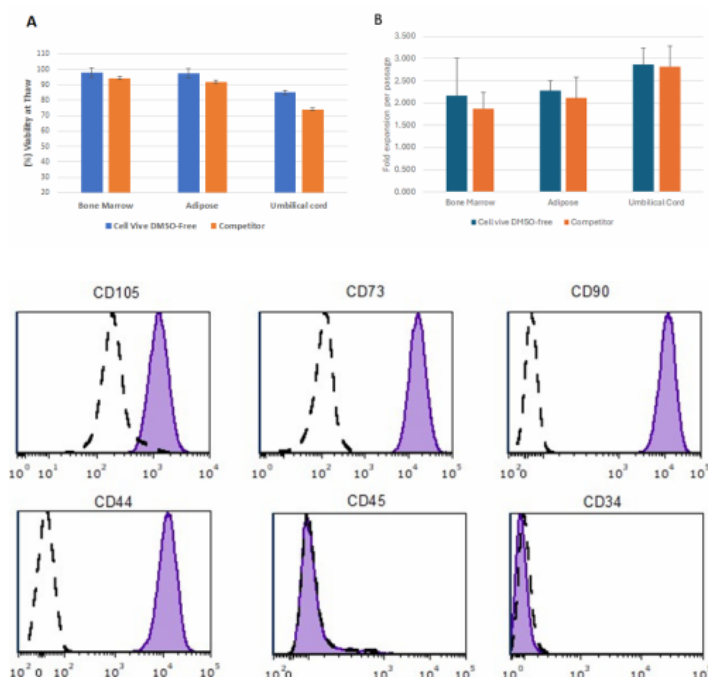
BioLegend Cell-Vive™ GMP Cell Culture products are for research use only. Suitable for *ex vivo* cell processing use. Not for injection or diagnostic or therapeutic use. Not for resale. BioLegend will not be held responsible for patent infringement or other violations that may occur with the use of our products.

### Antigen Details

Gene ID

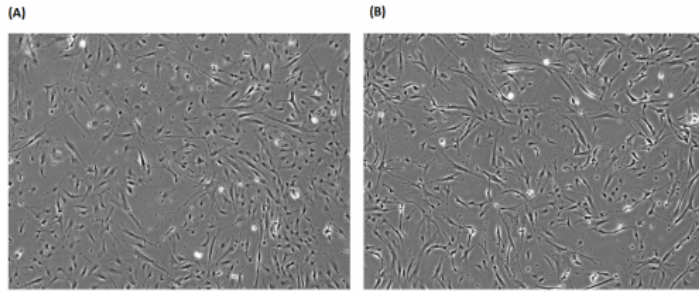
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### Product Data

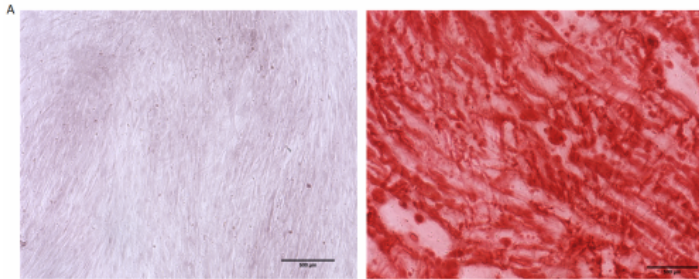


**Cell-Vive™ CD DMSO-Free Cryopreservation Solution, GMP preserves Human MSCs viability, and phenotype post-cryopreservation, at similar or higher levels than DMSO solution commonly used (Competitor).** After reaching 80% confluency, Human MSCs were detached and stored in liquid nitrogen for two days before thawing. **(A)** Viability at thaw was determined and cell expansion demonstrated through three passages **(B)**.

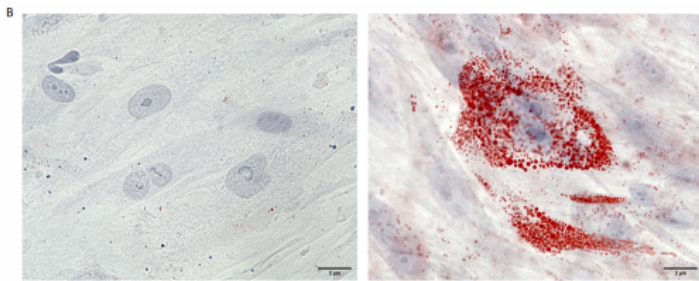
After three passages following cryopreservation and thaw, the MSC phenotype was analyzed by flow cytometry. Thawed and expanded MSCs maintained phenotype as demonstrated by CD105+, CD73+, CD90+, CD44+, CD45- and CD34- (filled histogram), isotype control (open histogram) markers.



**Cell-Vive™ CD DMSO-Free Cryopreservation Solution, GMP preserves Human MSCs morphology similar to other commonly used DMSO solution.** Human bone marrow-derived MSCs were frozen at 500,000 cells/mL in (A) Cell-Vive™ CD DMSO-Free Cryopreservation solution or a (B) DMSO solution (Competitor). Cells were stored in liquid nitrogen for two days, then thawed and plated at 5,000 cells/cm<sup>2</sup>. Attachment and morphology were observed four days after thawing. Images were taken at 4X magnification.



**Cell-Vive™ CD DMSO-Free Cryopreservation Solution, GMP preserves the differentiation capacity of Human Mesenchymal Stem Cells.** Human MSCs were stored in liquid nitrogen in the presence of Cell-Vive™ CD DMSO-Free Cryopreservation Solution, GMP for two days. MSCs were thawed and differentiated into (A) osteocytes for nineteen days or (B) adipocytes for twenty-one days with respective differentiation media (right) or cell culture media as a control (left). Osteocytes were stained with Alizarin red and adipocytes with Oil Red O.



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