HiFi™ Human Peripheral Blood Mononuclear Cells (H-PBMC), Pooled

PRODUCT INFORMATION

PRODUCT CODE: CL010

Product Warranty
HiFi™ cells are performance tested using HiMedia media and reagents. Users must ensure proper handling, storage and use of the product. **We DO NOT GUARANTEE the performance of the cells if protocols and products other than those mentioned in this sheet are used. We are not liable for any damage to the culture arising from improper storage or mishandling.**

Initial Handling upon Receipt
For any damage to the product upon receipt, report it immediately to the HiMedia marketing representative or technical team.

For any difficulty while handling or processing the cells please contact HiMedia technical team within a month after delivery.

022-61169797 / 8767552556 or atc@himedialabs.com

Check List

Cryopreserved Cells
Immediately transfer the vial to a liquid nitrogen tank upon receipt using forceps.

DO NOT hold the vial in hand.

DO NOT freeze the cells at -80°C.

Use sterile disposable tips and serological pipettes.

Avoid using autoclaved tips and serological pipettes.

Common Mistakes
Stem cells and primary cells are more sensitive and delicate as compared to the robust continuous cell lines hence, require extreme precautions while handling, subculturing and thawing.

Exposing the cryopreserved cells during thawing to warm temperatures (37°C) for more than 90 - 120 secs may lead to loss of cell viability.

Product Description and Contents
HiFi™ Human Peripheral Blood Mononuclear Cells (H-PBMC) are isolated from human peripheral blood. They are uncharacterized and contain normal mixture of lymphocytes, monocytes, macrophages and stem cells (round nucleated cells).

H-PBMC are frozen at density of less than 25 x 10⁶ (or 10 x 10⁶) cells per vial. Each lot of cells is pooled from three donors. Cells are maintained in an antibiotic-free environment prior to supply.

Source: Human
Tissue: Peripheral blood
Type of Cells: Mononuclear cells
Growth Characteristics: Suspension

<table>
<thead>
<tr>
<th>Products Required But Not Supplied</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>1. Growth medium</strong></td>
</tr>
<tr>
<td>Code</td>
</tr>
<tr>
<td>Growth medium RPMI-1640</td>
</tr>
<tr>
<td><strong>2. Media Supplements</strong></td>
</tr>
<tr>
<td>Fetal Bovine Serum (FBS)</td>
</tr>
<tr>
<td>RM10432</td>
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<tr>
<td>Antibiotic-Antimycotic Solution, 100X</td>
</tr>
<tr>
<td>Gentamicin-Amphotericin B Solution, 1000X</td>
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<tr>
<td><strong>3. Trypan Blue 0.5% solution</strong></td>
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<tr>
<td>TCL005</td>
</tr>
</tbody>
</table>
### Storage and Handling of Cryopreserved Cells

- Cryopreserved cells are supplied in liquid nitrogen dry vapour shipper (-150°C to -130°C).
- Upon receipt, immediately transfer the vial to the vapor phase of liquid nitrogen tank.
- Store it in the tank until further use. Cells must be processed at least in a BSL II hood.

#### Thawing Procedure

<table>
<thead>
<tr>
<th>Key Points to Remember</th>
<th>Time Required (approx.)</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Work in areas designated for use Human origin material Biosafety Level II (BSL II)</strong>&lt;br&gt;Wear protective clothing and gloves</td>
<td></td>
</tr>
<tr>
<td>Add 10ml of complete medium into conical bottom centrifuge tube (50 ml)</td>
<td>60 secs</td>
</tr>
<tr>
<td>Remove cryovial from the liquid nitrogen tank/shipper wearing appropriate protective gear</td>
<td>90-120 secs</td>
</tr>
<tr>
<td>Thaw the frozen cells in the vial partially by holding the vial in a water bath at 37°C</td>
<td>10 secs</td>
</tr>
<tr>
<td>Disinfect the vial by swabbing thoroughly with 70% isopropyl alcohol.</td>
<td></td>
</tr>
<tr>
<td>Add the cell suspension drop by drop to the conical bottom centrifuge tube containing the pre-warmed complete medium.</td>
<td>30-60 secs</td>
</tr>
<tr>
<td>Keep the conical bottom centrifuge tube swirling while addition of cell suspension.</td>
<td>30-60 secs</td>
</tr>
</tbody>
</table>

**Preparation of complete medium**<br>AL028A (450 ml) + (FBS 50 ml) + A002 (5 ml)<br>[Refer Technical Data Sheet (TDs) of respective medium]

**Make sure water bath is set at 37°C prior to beginning of the work**

**Thawing should be AS FAST AS POSSIBLE to minimize cell damage**

**DO NOT hold the vial in water bath for more than 90-120 secs**

**AVOID getting water upto the cap of the vial**

**Dropwise addition is required to prevent the cells from stress induced by exothermic reaction**
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<tr>
<td>Centrifuge at 1000 rpm for 10 min. Discard the supernatant and resuspend pellet in 20 ml medium in a conical bottom centrifuge tube</td>
<td>Mix Gently! Use cell suspension according to your experimental design</td>
</tr>
<tr>
<td>Mix gently to get uniform cell suspension</td>
<td>10-12 min</td>
</tr>
</tbody>
</table>

**Quality Control**

**Growth Performance Assay**

No. of viable cells/vial: NLT 25 X 10^6 cells/vial  
Percentage viability: NLT 80%

**Marker Analysis Assay By Flow Cytometry**

This data has been generated on 8 parameter 3 laser Partec CyFlow® Cube 8 Flow Cytometer and is specific for representative batch of H-PBMC.

- CD45+ : Positive  
- CD3+ : Lot specific expression  
- CD4+ : Lot specific expression  
- CD8+ : Lot specific expression  
- CD14+ : Lot specific expression

**Sterility Testing**

- Mycoplasma: Not detected  
- Bacteria, Fungi and Yeast: Not detected

**Virus Testing**

- HIV: Not detected  
- Hepatitis B Virus: Not detected  
- Hepatitis C Virus: Not detected

**Warning**

This product is intended for research use only. Not for animal, human therapeutic or diagnostic use.

Product contains human origin material and should be treated as potentially infectious. Even if the cells provided have been screened for viral and bacterial pathogens, human cells may harbour other known or unknown agents which might pose a health hazard. Universal handling precautions applicable to biological samples must be applied as recommended in the CDC-NIH manual.

**Disclaimer**

User must ensure suitability of the product(s) in their application prior to use. Products conform solely to the information contained in this and other related HiMedia™ publications. The information contained in this publication is based on our research and development work and is to the best of our knowledge true and accurate. HiMedia™ Laboratories Pvt. Ltd. reserves the right to make changes to specifications and information related to the products at any time. Products are not intended for human or animal diagnostic or therapeutic use but for laboratory, research or further manufacturing use only, unless otherwise specified. Statements contained herein should not be considered as a warranty of any kind, expressed or implied, and no liability is accepted for infringement of any patents.

HiMedia Laboratories Pvt. Ltd.  
A-516, Swastik Disha Business Park, Via Vadhaní Indl. Est. LBS Marg, Mumbai - 400 086, India.  
Customer Care No.: 022-61471919  |  Email: atc@himedialabs.com