STEM-CELLBANKER® DMSO Free GMP Grade

(Chemically defined cryopreservation solution)

For Research Use Only

Manufactured By

ZENOGEN PHARMA CO., LTD.

FDA DMF Registered

Cat#: 13926 (100mL), 13925 (20mL)

Storage Temperature: 2 to 8°C or below -20°C.

Expiry Date: 3 years from manufacturing date (see label)

Manufactured By: Zenogen Pharma Co., Ltd



Protocol:

Freezing

For optimum results, cells for cryopreservation should be in log phase of growth. Similar or other standard freezing protocols may be substituted.

- 1. Examine and make sure the cell culture is free of contamination, in healthy and at proper confluency.
- 2. Perform a cell count to determine the viability of cells.
- 3. Centrifuge at 1,000 2,000 rpm, 4°C for 3 to 5 minutes to gently pellet the cells. Remove the supernatant with an aspirator.
- 4. Gently suspend STEM-CELLBANKER® cryopreservation medium (1 mL for 5×10^5 5×10^6 cells).
- 5. Transfer 1 mL of the cell suspension to cryopreservation vial labeled with appropriate information (the cell line name, concentration, passage date etc.).
- 6. Place the vials directly in -80°C for storage.
- 7. **(OPTIONAL)** Transfer the frozen vials to a liquid nitrogen storage tank after the vials have been frozen for at least 24 hours.

IMPORTANT: Optimum protocol may change with the cell types.

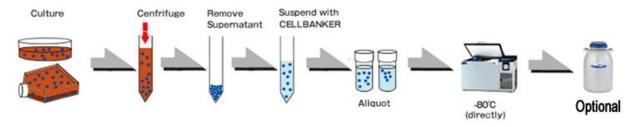
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Procedure for Use:



Thawing

- 1. Remove the cryopreservation vial from the freezer and quickly thaw cells in a 37°C shaking water bath or shake by hand.
- 2. Transfer the content to a centrifugation tube then immediately dilute and gently mix with 10mL of complete cell culture medium. Using CELLOTION® instead of complete culture medium will prevent adhesion of cells to the wall of the tube, increasing the recovery rate.
- 3. Centrifuge cells at 1,000 2,000 rpm, 4°C for 3 to 5 minutes. Remove the supernatant with an aspirator.
- 4. Gently resuspend the cells with appropriate volume of complete cell culture medium then plate in a culture flask or plate.
- 5. Continue the culture procedures according to standard protocols.

Cells Tested (Check website for updated list)

FDA Master File:

This product is registered with FDA Drug Master File. Please contact us at <u>orders@iwai-chem.com</u> or fill out the form online.

References:

Mao, X. & Zhao, S. Neuronal Differentiation from Mouse Embryonic Stem Cells In vitro. JoVE e61190 (2020) doi: 10.3791/61190.

Ueda, H. et al. Establishment of in vitro 3D spheroid cell cultivation from human gynecologic cancer tissues. STAR Protocols 2, 100354 (2021) doi: 10.1016/j.xpro.2021.100354.

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Yagishita, S. et al. Characterization of the large-scale Japanese patient-derived xenograft (J-PDX) library. Cancer Science 112, 2454–2466 (2021) doi: 10.1111/cas.14899.

Mikłosz, A. et al. Does TBC1D4 (AS160) or TBC1D1 Deficiency Affect the Expression of Fatty Acid Handling Proteins in the Adipocytes Differentiated from Human Adipose-Derived Mesenchymal Stem Cells (ADMSCs) Obtained from Subcutaneous and Visceral Fat Depots? Cells 10, (2021) doi: 10.3390/cells10061515.

Ueno, K. et al. Freezing of cell sheets using a 3D freezer produces high cell viability after thawing. Biochemistry and Biophysics Reports 28, 101169 (2021) doi: 10.1016/j.bbrep.2021.101169.

Disclaimer:

STEM-CELLBANKER® DMSO Free GMP grade is not itself a pharmaceutical. Therefore, no warranty, express or implied, as to the fitness and suitability of this product for any particular purpose and/or merchantability unless the use is intended for research.

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