PentaHibe®

PHARMACOSMOS

Protocol

DMSO-free cryopreservation of HPC(A) products using 16% Pentahibe® Base and 2% human albumin

The specific protocol for freezing cells depends on the cell type and cryopreservation solution used. For HPC(A) products, it is recommended to cryopreserve in 16% Pentahibe® Base supplemented with 2% human albumin[1].

Materials and reagents:

- 40% Pentahibe® Base
- 20% human albumin (HA)
- Ice

1 Preparation of cryopreservation solution

Formulation

On the day of use prepare a 2x concentration cryopreservation solution (32% Pentahibe® Base and 4% HA), e.g.,

1. Add 10 mL of 20% HA solution to a Pentahibe® Base vial, containing 40 mL solution
2. Mix gently by inverting the vial a few times
3. Keep the cryopreservation solution on ice until use

2 Cryopreservation procedure

Protocol (example)

1. Mix 2x cryopreservation solution 1:1 (v/v) with HPC(A) products using preferred storage container (cryobags or cryovials)
2. Mix gently by hand by carefully inverting the container a few times
3. Incubate on ice for 15 min
4. Cells are frozen using a controlled rate freezer, e.g. utilizing gradient-based cooling (start temp 4°C, -1°C/min drop to 0°C, -2°C/min drop to -45°C, and -5°C/min drop to -100°C), followed by storing cells in a liquid nitrogen container

3 Thawning

1. Thaw cells rapidly in a 37°C water bath. Thawing should be done gently by swirling the sample until all visible ice has just melted
2. The cells are now ready to be processed as is, or can be diluted with an appropriate culture medium pre-warmed to 20-37°C at a dilution ratio of 1:10 (sample to culture medium)

[1] Svalgaard JD et al. Pentaisomaltose, an alternative to DMSO. Engraftment of cryopreserved human CD34+ cells in immunodeficient NSG mice. Cell Transplantation 2018, Vol. 27(9):1407-1412.

We are here to answer your questions

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