ImmunoPure™ HD

Normal Human CD4+ Helper T Cells

Genlantis
A division of Gene Therapy Systems, Inc.

Catalog #	Description/Content	Amount
PBMC003A	Normal Human CD4+ Helper T Cells, Cryopreserved	10.0 x 10^6 cells

Shipping and Storage:	Cells are shipped on dry ice. **For maximum viability, it is best to use cells as soon as possible**. For short term (3 weeks or less) store cells at -80ºC; for longer term, store cells in liquid nitrogen (-170ºC). NOTE: viability cannot be guaranteed following -170ºC storage or beyond one week at -80ºC.

Related Products	Catalog Numbers
Normal Human Peripheral Blood Mononuclear Cells	PBMC000 (5.0x10^6); PBMC001 (25.0x10^6); PBMC002 (75.0x10^6).
Normal Human CD8+ Cytotoxic T cells	PBMC004A (10.0x10^6).
Normal Human CD14+ Monocytes	PBMC005A (10.0x10^6).
Normal Human CD19+ B Cells	PBMC006A (10.0x10^6).
Normal Human CD56+ Natural Killer Cells	PBMC007A (10.0x10^6).
Normal Human Dendritic Cells	PBMC008A (0.5x10^6).

INTRODUCTION

Normal Human CD4+ Helper T cells are a sub-group of lymphocytes that play a crucial role in establishing and maximizing the capabilities of the immune system. These cells are unusual in that they have no cytotoxic or phagocytic activity; they cannot kill infected host cells (also known as somatic cells) or pathogens. CD4+ T cells are essential in determining B cell antibody class switching, in the activation and growth of cytotoxic T cells, and in maximizing bactericidal activity of phagocytes such as macrophages. It is this diversity in function and their role in influencing other cells that gives T helper cells their name. The ImmunoPure HD Human CD4+ Helper T Cells are isolated from fresh human adult peripheral blood of healthy donors at IRB-approved and FDA-licensed blood banks, using apheresis and immunomagnetic cell separation techniques.

MATERIALS AND METHODS

A. General Medium Requirement

RPMI 1640 medium + 10% (v/v) FBS, 2 mM glutamine, 1% (v/v) nonessential amino acids, 1% (v/v) sodium pyruvate, 50 U/ml penicillin, 50 mg/ml streptomycin, and 50 mg/ml kanamycin.

B. Thawing and Culturing Cells

1. Prepare a 37ºC water bath to temperature.
2. Keep all samples frozen until the bath is ready.
3. Place the vials into the water bath, being careful not to submerge below the junction between the lid and the vial.
4. Place vial of cells in 37°C water bath and agitate until thawed. It is important to thaw the cells quickly; do **NOT** allow thawed cells to remain in freezing media any longer than necessary.
5. When only a small amount of ice remains, remove the vials and dry with a lab tissue. Clean the top of the vial with a lab tissue moistened with 70% alcohol; **avoid wiping away the labeling.**
6. Within about 30 sec., slowly add one milliliter of medium (containing serum) to the thawed cells.
7. Slowly add thawed cells to 8 ml of medium containing serum. Invert tube 2 or 3 times to mix or mix gently by pipetting up and down several times.
8. Centrifuge for 5 min at 400 x g.
9. Aspirate or decant the supernatant and gently resuspend the cell pellet in 10 ml of medium.
10. Remove an aliquot for cell count and proceed with experimental manipulations.
11. Culture the cells in RPMI 1640 medium supplemented with 10% (v/v) FBS, 2 mM glutamine, 1% (v/v) nonessential amino acids, 1% (v/v) sodium pyruvate, 50 U/ml penicillin, 50 mg/ml streptomycin, and 50 mg/ml kanamycin at a density of 1-2 million cells/ml.
    NOTE: The cell suspension may form clumps after standing at room temperature. This can be avoided by preparing and using the cells promptly or by adding DNase to the suspension at a final concentration of 10 units per ml.

C. References

1. CXCR5 Expressing Human Central Memory CD4 T Cells and Their Relevance for Humoral Immune Responses. Nina Chevalier, David Jarrossay, Edwin Ho, Danielle T. Avery, Cindy S. Ma, Di Yu, Federica Sallusto, Stuart G. Tangye and Charles R. Mackay. *J Immunol 2011; 186; 5556-5568.*

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