GMP Human T Cell Media

Media for Human T Cell Culture

Catalog Number: CCM038-GMP-1L
CCM038-GMP-1B

PRODUCT DESCRIPTION

GMP Human T Cell Media is Xeno-free and does not contain non-human animal-derived components. GMP Human T Cell Media is formulated and optimized for the *ex vivo* culture of human T lymphocytes for preclinical or clinical use. This product does not contain antibiotics or phenol red. This media can be used to culture primary T cells, immortal T cell lines and T cell clones, the stimulation of peripheral blood lymphocytes (PBLs), and study of mixed leukocyte reactions (MLR).

INTENDED USE

GMP Human T Cell Media is a versatile media compatible with various T cell activation methods (any CD3/CD28 activation reagent can be utilized), platforms (G-Rex, cell culture bags, flasks, and plates) and different cytokine/growth factor. This media supports the expansion of T cells in protocols using IL-7 and IL-15, or IL-2 cytokines of various concentrations. Supplements for the *ex vivo* culture of human T lymphocytes. The cytokine/growth factor combination used depends upon the experimental design of each researcher.

MATERIALS PROVIDED & STORAGE

PART	CATALOG #	SIZE	STORAGE & STABILITY
GMP Human T Cell Media	CCM038-GMP-1L	1000 mL, Bottle	Upon receipt, store at 2-8 °C and **protect from light** until the use by date on the CofA.
GMP Human T Cell Media	CCM038-GMP-1B	1000 mL, Bag	

PRECAUTIONS

The human origin-derived components used in this product have been derived from human plasma, which has been tested at the donor level and found to be negative for antibodies to HIV-1/2, hepatitis B surface antigen (HBsAg), and hepatitis C virus (HCV). Safe laboratory procedures should be followed, and protective clothing should be worn when handling this media. The acute and chronic effects of over-exposure to this media are unknown.

LIMITATIONS

- For preclinical or *ex vivo* clinical use.
- This reagent should not be used beyond the use by date indicated on the label.
- Results may vary due to variations among primary T lymphocyte populations derived from different donors.

OTHER MATERIALS REQUIRED

- Recombinant Human IL-7 GMP Protein, CF Catalog #, BT-007-GMP or ProPak™ Recombinant Human IL-7 GMP Protein, CF Catalog #, PPK-007-GMP
- Recombinant Human IL-15 GMP Protein, CF Catalog #, BT-015-GMP or ProPak™ Recombinant Human IL-15 GMP Protein, CF Catalog #, PPK-015-GMP
- Recombinant Human IL-2 GMP Protein, CF *(alternatively)* Catalog #, BT-002-GMP
- Human CD3 GMP Antibody Catalog #, MAB11411-GMP
- Human CD28 GMP Antibody Catalog #, MAB11412-GMP
- G-Rex® Bioreactors, Wilson Wolf™
- Human AB Serum

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PROCEDURE FOR THE EX VIVO CULTURE OF HUMAN T CELLS

The protocol describes the expansion of Human T Cells using GMP Human T Cell Media.
Note: *The activation and cytokine/growth factor combinations used with this media should be optimized by application or experimental protocol.*

RECOMMENDED REAGENT PREPARATION FOR COMPLETE GMP HUMAN T CELL MEDIA

Bottles (CCM038-GMP-1L) - Aliquot media and add 10 ng/mL rhIL-7 and 10 ng/mL rhIL-15 (or alternatively, 10 ng/mL rhIL-2). Complete media is stable for 2 weeks at 2-8 °C. Store protected from light.
Optional: 5% Human AB Serum is highly recommended for optimal culture conditions.

Bags (CCM038-GMP-1B) - Cytokine addition can be achieved through the weldable PVC line or the needleless injection port. Read User Manual: Addition of ProPak™ Cytokines for Closed Process Manufacturing for closed system addition with ProPaks. Complete media (cytokines and/or human AB serum added) is stable for 2 weeks at 2-8 °C. Store protected from light.
Optional: 5% Human AB Serum is highly recommended for optimal culture conditions.

RECOMMENDED PROTOCOL (G-Rex®6M Well Plate)

1. Isolate T cells or thaw isolated T cells using desired protocol.
2. Plate T cells at 0.5 x 10^6/cm^2 in 1 mL/ cm^2 of complete GMP Human T cell Media in G-Rex bioreactor.
   a. Example: 5 x 10^6 cells in 10 mL media in each well of a G-Rex 6M Well Plate.
3. Activate cells with CD3/CD28 activation reagent of choice using manufacturer's specifications.
4. *Optional: Perform gene editing by either lentivirus transduction or TcBuster transposition*
   a. Transduction: 24 hours after activation, carefully mix the cell culture to break up the complexes and count the cell numbers. Apply lentivirus (recommend MOI of 5), and transduction enhancer if desired, to cells and gently mix. Recommended to omit presence of Human AB serum in media during thaw, activation, and transduction of T cells. Human AB serum prior to and during transduction.
   b. TcBuster Transposition: 42-48 hours after activation, introduce CAR/TCR construct via electroporation with platform of choice using TcBuster-M transposase and transposon (see TcBuster electroporation protocols for detailed information about platform specific recommendations).
5. Approximately 24 hours after transduction or 48 hours after TcBuster transposition, bring volume of G-Rex bioreactor to 10 mL/cm^2 with fresh complete media.
6. Expand cells in 37 °C incubator without manipulation.
7. Recommend harvest timing 7-9 days of culture or when cell confluency exceeds 30 x 10^6 /cm^2.
8. Additional T cell activation and expansion protocols using GMP Human T Cell Media:
   - T Cell CD3 Immobilized, CD28 Soluble
   - T Cell Bead-Conjugated
   - PBMCs Soluble and Bead-Conjugated

END USER TERMS OF USE OF PRODUCT

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