

# ArtMedia<sup>®</sup> Human T Cell Serum-Free Medium (GMP Grade, with phenolic red)

Please read the datasheet carefully prior to use.

Cat. No. MT101

Version No. Version 1.0

Storage at 2-8°C for one year

## Description

ArtMedia<sup>®</sup> Human T Cell Serum-Free Medium (GMP Grade) is a serum-free, xeno-free and chemically defined medium for the culture of human T lymphocytes. With the combined effect of antibodies CD3/CD28 and IL-2, the medium can support the rapid expansion of T cells of peripheral blood mononuclear cells (PBMCs) and maintain their potential.

## Features

- Serum-free, xeno-free and chemically defined
- With phenolic red, without antibiotics
- Supports rapid expansion and high-density culture of T cells
- A variety of T cell activation methods (antibody soluble method, antibody coating method, antibody conjugated magnetic bead method) is optional.
- Produced and managed in accordance with GMP standards

## Kit Contents

Component	MT101-01
ArtMedia <sup>®</sup> Human T Cell Serum-Free Medium (GMP Grade)	1000 ml

## Procedures

### 1. Preparation of serum-free complete medium for T cells

Add IL-2 (TransGen, Cat: PM101) with a final concentration of 100~500 IU/ml to ArtMedia<sup>®</sup> Human T Cell Serum-Free Medium (GMP Grade) to prepare a serum-free complete medium for T cells (add IL-2 at the appropriate concentration according to different experimental conditions, 200 IU/ml is recommended). Use it right after it was ready.

### 2. Coating of the culture plate (optional)

- (1) Prepare antibody-coated plates for T cell activation: dilute Anti-Human CD3 mAb (TransGen, Cat: HM101) and Anti-Human CD28 mAb (TransGen, Cat: HM102) with PBS to 0.5~5 µg/ml, then add to the plate ready to be coated and ensure that the liquid covers the entire plate bottom (select the antibody concentration with the best activation effect according to different experimental conditions, the recommended final concentration is 1 µg/ml).
- (2) Seal with parafilm and incubate overnight at 2~8°C.
- (3) Plate can be seeded with cells after 24 hours, or continue to be incubated at 2~8°C and use within 3 days

### 3. Activation and culture of T cells

- (1) Place the culture plate coated with CD3/CD28 antibody in a incubator for 30 minutes to equilibrate it to 37°C.
- (2) Fresh PBMCs or resuscitated PBMCs are isolated by Human Peripheral Blood Lymphocyte Separation Solution (TransGen, Cat: FB102).
- (3) Resuspend and wash cells with T cell serum-free medium, count cells, and record cell viability and viable cell number.
- (4) Centrifuge at 300×g for 5 minutes and discard the supernatant. Resuspend the cells with T cells at 0.5~1×10<sup>6</sup> live cells/ml in serum-free complete medium, supplemented with IL-2.



- (5) Discard the antibody mixture in the coated plate, transfer PBMC according to the corresponding volume of the plate, and incubate culture plate in 5% CO<sub>2</sub> incubator (a variety of protocols may be used for activating T cells according to experimental conditions, including antibody soluble method or CD3/CD28 antibody conjugated magnetic beads method).
- (6) There is no need to change or supplement the medium for the first 2 days after seeding. And keep static cultivation.
- (7) On day 3, record the viability and viable cell number. According to the counting results, supplement with fresh T cell serum-free complete medium, adjust the cell density to  $0.5 \times 10^6$  live cells/ml, or change the medium completely according to experimental conditions.
- (8) On the 5th day of cell activation, the T cells with static cultivation can be transferred to a shaker or culture bag. Count samples every two days, supplement with fresh T cell serum-free complete medium and adjust the cell density to  $0.5 \times 10^6$  viable cells/ml.
- (9) On the 9th~11th day, harvest the cells according to the experiment.

#### Notes

- Please prepare fresh T cell serum-free complete medium supplemented with IL-2 before each experiment.
- T cell serum-free medium can be used with a variety of T cell activation methods. Please explore the best activation scheme according to the experimental conditions.
- If a longer period of culture is required, a second activation and expansion culture can be performed.
- Cell proliferation and subtype proportions will vary due to sample variability.

**For research use only, not for clinical diagnosis.**

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