CTS™ OpTmizer™ T-Cell Expansion SFM

Catalog Numbers A1048501, A1048503, A3705001, A3705003

Pub. No. MAN0007325 Rev. 4.0

WARNING! Read the Safety Data Sheets (SDSs) and follow the handling instructions. Wear appropriate protective eyewear, clothing, and gloves. Safety Data Sheets (SDSs) are available from **thermofisher.com/support**.

Product description

Gibco™ CTS™ OpTmizer™ T-Cell Expansion SFM has been developed for the growth and expansion of human T lymphocytes. CTS™ OpTmizer™ T-Cell Expansion Medium is a complete serum-free, xeno-free 1X medium consisting of CTS™ OpTmizer™ T-Cell Expansion Basal Medium with the addition of CTS™ OpTmizer™ T-Cell Expansion Supplement. Each container is a sterile filtered single-use container.

Contents and storage

CTS™ OpTmizer™ T-Cell Expansion SFM is sold as a complete kit. The components are not sold separately.

Contents	Amount	Storage	Shelf life[1]
CTS™ OpTmizer™ T-Cell Expansion SFM, Cat. No. A1048501			
CTS™ OpTmizer™ T-Cell Expansion Basal Medium	1000 mL (Bottle)	2°C to 8°C. Protect from light.	18 months
CTS™ OpTmizer™ T-Cell Expansion Supplement	26 mL		
CTS™ OpTmizer™ T-Cell Expansion SFM, Cat. No. A1048503			
CTS™ OpTmizer™ T-Cell Expansion Basal Medium	1 L (Media Bag)	2°C to 8°C. Protect from light.	18 months
CTS™ OpTmizer™ T-Cell Expansion Supplement	26 mL		
CTS™ OpTmizer™ T-Cell Expansion SFM, no phenol red, Cat. No. A3705001			
CTS™ OpTmizer™ T-Cell Expansion Basal Medium	1000 mL (Bottle)	2°C to 8°C. Protect from light.	18 months
CTS™ OpTmizer™ T-Cell Expansion Supplement	26 mL		
CTS™ OpTmizer™ T-Cell Expansion SFM, no phenol red, Cat. No. A3705003			
CTS™ OpTmizer™ T-Cell Expansion Basal Medium	1 L (Media Bag)	2°C to 8°C. Protect from light.	18 months
CTS™ OpTmizer™ T-Cell Expansion Supplement	26 mL		

[1] Shelf-Life duration is determined from Date of Manufacture.

Safety information

Human origin materials are non-reactive (donor level) for anti-HIV 1 & 2, anti-HCV, and HBsAg. Handle in accordance with established bio-safety practices.

Culture conditions

Media: Complete CTS™ OpTmizer™ T-Cell Expansion SFM

Cells: Peripheral Blood Mononuclear Cells (PBMC)

Culture type: Suspension

Culture vessels: T-Flasks or Xuri™ Cellbag™ Bioreactor

Temperature range: 36°C to 38°C

Incubator atmosphere: Humidified atmosphere of 5% CO_2 in air. Ensure proper gas exchange and minimize exposure of cultures to light.

Procedural guidelines

- Do not freeze CTS™ OpTmizer™ T-Cell Expansion Supplement.
- Foaming may occur during shipment of the supplement, but will not impact performance of the product.
- Supports high density $CD3^+$ T-cell cultures (e.g., > 3×10^6 cells/mL) in static and (e.g., $>2 \times 10^7$ cells/mL) WAVE Cellbag™ cultures.

Prepare media

CTS™ OpTmizer™ T-Cell Expansion Basal Medium requires supplementation with CTS™ OpTmizer™ T-Cell Expansion Supplement, and L-glutamine.

Note: To prepare complete 1X medium in the media bag, use a needle syringe to aseptically inject the supplement(s) into the media bag via the self sealing injection site.

1. Place the CTS™ OpTmizer™ T-Cell Expansion Basal Medium, CTS™ OpTmizer™ T-Cell Expansion Supplement, and thawed L-glutamine (200 mM) under a sterile laminar flow hood.
2. Add 26 mL CTS™ OpTmizer™ T-Cell Expansion Supplement to 1 L CTS™ OpTmizer™ T-Cell Expansion Basal Medium.

   Discard pipette.
3. Using a new sterile pipette, remove 10 mL of L-glutamine (200 mM) and add to 1 L of CTS™ OpTmizer™ T-Cell Expansion Basal Medium.

   Discard pipette.
4. Replace the caps tightly and swirl gently to mix the complete CTS™ OpTmizer™ T-Cell Expansion SFM.
5. Medium can be further supplemented with cytokines and/ or antibiotics if desired following steps 1–3.
6. Complete 1X CTS™ OpTmizer™ T-Cell Expansion SFM may be supplemented with cytokines such as IL-2 to support T-cell expansion. It is recommended to use 100–200 IU/mL of IL-2 for standard T cell expansion. The amount of IL-2 used may vary depending on experimental conditions.
7. If desired, antibiotics can be used. It is recommended to use Gentamicin at 10–50 µg/mL or Penicillin-Streptomycin.

   Note: CTS™ OpTmizer™ T-Cell Expansion SFM is designed to support T-cell cultures without the addition of human serum. If required, 2% heat-inactivated human serum may be added to the medium to enhance viability and expansion. Alternatively, CTS™ Immune Cell SR may be used in place of human serum. The use of serum or CTS™ Immune Cell SR and the amount required, should be determined empirically depending on the specific T-cell culture application.

   Once the complete CTS™ OpTmizer™ T-Cell Expansion SFM (basal medium with supplement, and L-glutamine) is prepared in accordance with our instructions, it must be stored in the dark at 2°C to 8°C and used within four weeks of supplementing to be covered by our warranty.

Culture T-cells

General guideline for all static T-cell cultures, regardless of vessel. For high-density culture in bioreactors, such as WAVE Cellbag™ Bioreactor, optimal procedures should be determined empirically by the investigator.

1. Prepare fresh peripheral blood mononuclear cells (PBMCs) or rapidly thaw (<1 minute) frozen vials of PBMCs cells in a 37°C water bath according to standard PBMC thawing protocols.
2. Wash cells with DPBS, no calcium, no magnesium, with 5% heat-inactivated FBS or heat-inactivated human pooled Type AB serum according to the applications, if desired or required.
3. Determine total viable cell density and cell viability using Countess™ II Automated Cell Counter.
4. Centrifuge cells at 200 × g for 5–10 minutes and remove wash buffer.
5. Resuspend PBMCs at $0.5–1 \times 10^6$ cells/mL in 1X complete CTS™ OpTmizer™ T-Cell Expansion SFM, supplemented with cytokines if used at culture initiation.
6. Transfer the required number of cells to the appropriate tissue culture vessel.

   Note: A variety of protocols may be used for activating T-cells for subsequent expansion, including adding stimulatory antibodies or antigen presenting cells. Similarly, for either small or the large scale T-cell expansion, cells can be isolated, activated and expanded with CTS™ Dynabeads™ CD3/CD28 according to instructions in the product insert.
7. Incubate the culture vessel at 37°C in a humidified atmosphere of 5% CO_2 in air.
8. Feed and maintain cells at desired concentrations while cells are in log phase growth.

   To maintain log phase growth, it may be preferable to split cells to achieve a density of $0.5–1 \times 10^6$ T-cells/mL whenever cell density gets above 1×10^6 cells/mL (e.g. 2×10^6 cells/mL would be split 1:4 to continue culture at 0.5×10^6 cells/mL).

   Note: For optimal gas exchange in static plate cultures it is recommended that medium depth not exceed 1–1.2 cm.

Related products

Unless otherwise indicated, all materials are available through **thermofisher.com**.

Item	Source
CTS™ DPBS without calcium chloride, without magnesium chloride	A12856
L-Glutamine	25030
CTS™ GlutaMAX™-I Supplement	A1286001
AB-Human Serum	34005
Fetal bovine serum, certified, heat inactivated, US origin	10082
Gentamicin (50 mg/mL)	15750
Penicillin-Streptomycin	15070
Countess™ II Automated Cell Counter	AMQAX1000
CTS™ Dynabeads™ CD3/CD28	40203D
CTS™ DynaMag™ Magnet	12102
Dynabeads™ Human T-Expander CD3/CD28	11141D

Limited product warranty

Life Technologies Corporation and/or its affiliate(s) warrant their products as set forth in the Life Technologies' General Terms and Conditions of Sale at **www.thermofisher.com/us/en/home/global/terms-and-conditions.html**. If you have any questions, please contact Life Technologies at **www.thermofisher.com/support**.

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For descriptions of symbols on product labels or product documents, go to **thermofisher.com/symbols-definition**.