## ImmunoCult™-XF

# cGMP, serum-free, and xeno-free medium for the expansion of human T cells

Catalog #100-0956 500 mL



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## **Product Description**

ImmunoCult™-XF is a serum-free and xeno-free medium optimized for the in vitro culture and expansion of human T cells isolated from peripheral blood. Recombinant cytokines, required for the optimal growth and expansion of T cells, have not been added to ImmunoCult™-XF. This allows users the flexibility to prepare a medium that meets their requirements.

- No need to supplement the medium with serum
- Supports robust T cell expansion with high viability after 10 12 days of culture
- Expanded T cells are able to produce cytokines including IFN-gamma and IL-4 upon restimulation
- Use with ImmunoCult™ Human T Cell Activators (Catalog #10970 and #10971) for bead-free activation of T cells

ImmunoCult<sup>TM</sup>-XF is manufactured and tested following relevant cGMP guidelines under a certified quality management system. For additional information on quality at stemcell, refer to www.stemcell.com/compliance.

### **Properties**

Storage: Store at 2 - 8°C. Do not freeze.

Shelf Life: Stable until expiry date (EXP) on label.

This product contains material derived from human plasma manufactured under cGMP regulations. Donors have been tested and found negative for HIV-1 and -2, hepatitis B, and hepatits C prior to donation. However, this product should be considered potentially infectious and treated in accordance with universal handling precautions.

#### Directions for Use

The following protocol is for the expansion of activated human T cells using ImmunoCult™-XF. Depending on the experimental objectives, the protocol may need to be optimized (e.g. cell seeding density, culture duration, or choice of cytokine selection).

- Isolate human T cells from fresh or previously frozen peripheral blood mononuclear cells, or leukapheresis samples, using one of the following EasySep™ kits:
  - EasySep™ Release Human CD3 Positive Selection Kit (Catalog #17751)
  - EasySep™ Human T Cell Enrichment Kit (Catalog #19051)
  - EasySep™ Human T Cell Isolation Kit (Catalog #17951)

NOTE: Isolated T cells can be cryopreserved using CryoStor® CS5 (Catalog #07933) or CryoStor® CS10 (Catalog #07930) and stored at -135°C.

- 2. Day 0:
  - a. Prepare fresh complete ImmunoCult™-XF as follows:
    - Add cytokines (e.g. Human Recombinant IL-2; Catalog #78036/78145) to ImmunoCult™-XF. Mix thoroughly.
  - NOTE: Complete ImmunoCult™-XF must be prepared fresh on each day of use.
  - b. Seed viable human T cells (prepared in step 1) in fresh complete ImmunoCult™-XF (as prepared in step 2a) at 1 x 10^6 cells/mL.
- 3. To activate T cells, add 25 µL/mL of ImmunoCult™ Human CD3/CD28/CD2 T Cell Activator (Catalog #10970) or ImmunoCult™ Human CD3/CD28 T Cell Activator (Catalog #10971) to the cell suspension. Incubate cells at 37°C and 5% CO₂ for 3 days.
- 4. Day 3: Mix the cell suspension thoroughly and perform a viable cell count. Adjust the viable cell density to  $\sim 1$  2.5 x 10^5 cells/mL by adding fresh complete ImmunoCult<sup>TM</sup>-XF. Incubate at 37°C and 5% CO<sub>2</sub> for 2 days.
- 5. Day 5: Mix the cell suspension thoroughly and perform a viable cell count. Adjust the viable cell density to ~1 3 x 10^5 cells/mL by adding fresh complete ImmunoCult<sup>TM</sup>-XF. Incubate at 37°C and 5% CO<sub>2</sub> for 2 days.
- Day 7: Mix the cell suspension thoroughly and perform a viable cell count. Adjust the viable cell density to ~3 6 x 10<sup>5</sup> cells/mL) by adding fresh complete ImmunoCult™-XF. Incubate at 37°C and 5% CO₂ for 3 days.
- 7. Day 10: Harvest cells if the desired cell number is achieved.



OPTIONAL: Perform a viable cell count and maintain cell density at  $0.5 - 1 \times 10^6$  cells/mL by adding fresh complete ImmunoCult<sup>TM</sup>-XF. Incubate at 37°C and 5% CO<sub>2</sub> for 2 days, then harvest cells.

### **Notes and Tips**

For longer-term expansion (> 12 days) of human T cells:

- 1. On Day 7, harvest and resuspend the expanded T cells at 1 x 10^6 cells/mL in fresh complete ImmunoCult<sup>TM</sup>-XF.
- Restimulate by adding 25 µL/mL of ImmunoCult™ Human CD3/CD28/CD2 T Cell Activator or ImmunoCult™ Human CD3/CD28 T Cell
  Activator.
- 3. Incubate at 37°C and 5% CO<sub>2</sub>. Every 2 3 days, adjust cell density by adding fresh complete ImmunoCult<sup>TM</sup>-XF.
- ImmunoCult<sup>TM</sup>-XF can be supplemented with IL-7 (e.g. Catalog #78053) and/or IL-15 (e.g. Catalog #78031).
- Ensure to add fresh complete medium every 2 3 days of culture; do not wait more than 3 days between medium additions.

#### Related Products

For related products, including specialized culture and storage media, supplements, antibodies, cytokines, and small molecules, visit www.stemcell.com/TCellEngineering or contact us at techsupport@stemcell.com.

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