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Product Sheet

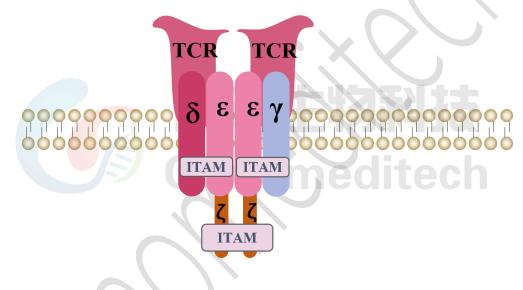
Cynomolgus_CD3 HEK-293 Cell Line

Catalog number: GM-C30879

Version 3.3.1.201016

The CD3 complex, also known as the T3 complex, is a multimeric protein complex composed of four different polypeptide chains: epsilon (ϵ), gamma (γ), delta (δ), and zeta (ζ). These chains assemble and function as three pairs of dimers: $\epsilon \gamma$, $\epsilon \delta$, and $\zeta \zeta$. The CD3 protein complex is a defining feature of the T-cell lineage, thus anti-CD3 antibodies can be effectively used as markers for T cells.

Cynomolgus_CD3 HEK-293 Cell Line is a clonal stable HEK-293 cell line constitutively expressing Cynomolgus CD3 complex.





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Specifications

Quantity 5E6 Cells per vial,1 mL

Product Format 1 vial of frozen cells

Shipping Shipped on dry ice

Storage Conditions Liquid nitrogen immediately upon receipt

Recovery Medium DMEM+10% FBS+1% P.S

Growth medium DMEM+10% FBS+1% P.S+400 μg/mL G418+125 μg/mL Hygromycin

Note None

Freezing Medium 90% FBS+10% DMSO

Growth properties Adherent

Growth Conditions 37°C, 5% CO₂

Mycoplasma Testing The cell line has been screened to confirm the absence of Mycoplasma species.

Safety considerations Biosafety Level 2

Note It is recommended to expand the cell culture and store a minimum of 10 vials at an early

passage for potential future use.

Materials

Reagent	Manufacturer/Catalogue No.
Hygromycin	Genomeditech/GM-040403
G418	Genomeditech/GM-040402
Pen/Strep	Thermo/15140-122
Fetal Bovine Serum	Cegrogen biotech/A0500-3010
DMEM	Gibco/C11995500BT
Anti-CD3 hIgG1 Antibody(CH2527)	Genomeditech/GM-33037AB



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Figures

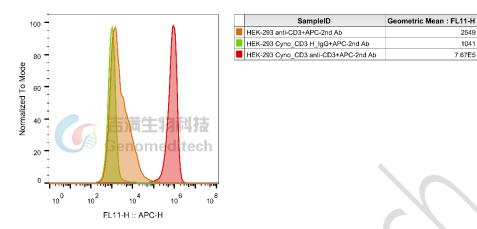


Figure 1 | Cynomolgus_CD3 HEK-293 Cell Line was determined by flow cytometry using Anti-CD3 hIgG1 Antibody(CH2527) (Cat. GM-33037AB).

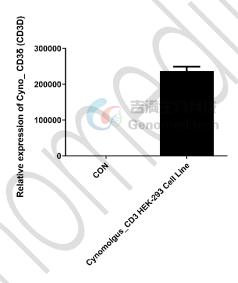


Figure 2 | The mRNA expression levels of Cynomolgus_CD3δ(CD3D) in the Cynomolgus_CD3 HEK-293 Cell Line (Cat. GM-C30879) were determined by RT-qPCR.



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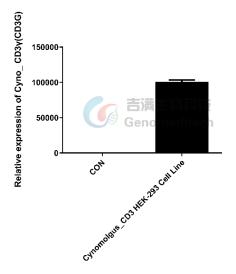


Figure 3 | The mRNA expression levels of Cynomolgus_CD3 γ (CD3G) in the Cynomolgus_CD3 HEK-293 Cell Line (Cat. GM-C30879) were determined by RT-qPCR.

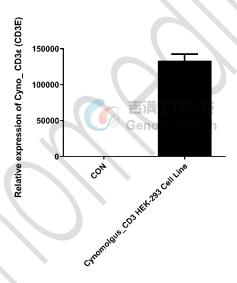


Figure 4 | The mRNA expression levels of Cynomolgus_CD3 ϵ (CD3E) in the Cynomolgus_CD3 HEK-293 Cell Line (Cat. GM-C30879) were determined by RT-qPCR.



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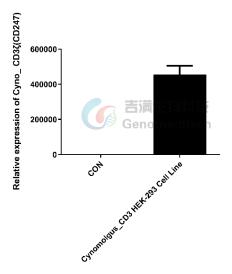


Figure 5 | The mRNA expression levels of Cynomolgus_CD3ζ(CD247) in the Cynomolgus_CD3 HEK-293 Cell Line (Cat. GM-C30879) were determined by RT-qPCR.

Cell Recovery

Recovery Medium: DMEM+10% FBS+1% P.S

To insure the highest level of viability, thaw the vial and initiate the culture as soon as possible upon receipt. If upon arrival, continued storage of the frozen culture is necessary, it should be stored in liquid nitrogen vapor phase and not at -70°C. Storage at -70°C will result in loss of viability.

- Thaw the vial by gentle agitation in a 37°C water bath. To reduce the possibility of contamination, keep the O-ring and cap out of the water. Thawing should be rapid (approximately 2 - 3 minutes).
- Remove the vial from the water bath as soon as the contents are thawed, and decontaminate by dipping in or spraying with 70% ethanol. All of the operations from this point on should be carried out under strict aseptic conditions.
- Transfer the vial contents to a centrifuge tube containing 5.0 mL complete culture medium and spin at approximately 176 x g for 5 minutes. Discard supernatant.
- Resuspend cell pellet with the recommended recovery medium. And dispense into appropriate culture dishes.
- Incubate the culture at 37°C in a suitable incubator. A 5% CO₂ in air atmosphere is recommended if using the medium described on this product sheet.

Cell Freezing

Freezing Medium: 90% FBS+10% DMSO

- Centrifuge at 176 x g for 3 minutes to collect cells. a)
- Resuspend the cells in pre-cooled freezing medium and adjust the cell density to 5E6 cells/mL. b)
- Aliquot 1 mL into each vial. c)



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d) Place the vial in a controlled-rate freezing container and store at -80°C for at least 1 day, then transfer to liquid nitrogen as soon as possible.

Cell passage

Growth medium: DMEM+10% FBS+1% P.S+400 µg/mL G418+125 µg/mL Hygromycin

For the first 1 to 2 passages post-resuscitation, use the recovery medium. Once the cells have stabilized, switch to a growth medium.

- a) Remove and discard culture medium.
- b) Briefly rinse the cell layer with PBS to remove all traces of serum that contains trypsin inhibitor.
- c) Add 1.0 mL of 0.25% (w/v) Trypsin-EDTA solution to dish and observe cells under an inverted microscope until cell layer is dispersed (usually within 30 to 60 seconds at 37°C).
- d) Note: To avoid clumping do not agitate the cells by hitting or shaking the flask while waiting for the cells to detach.

 Cells that are difficult to detach may be placed at 37°C to facilitate dispersal.
- e) Add 2.0 mL of growth medium to mix well and aspirate cells by gently pipetting.
- f) After centrifugation, resuspend the pellet and add appropriate aliquots of the cell suspension to new culture vessels.
- g) Incubate cultures at 37°C.

Subcultivation Ratio: A subcultivation ratio of 1:3 - 1:4 is recommended

Medium Renewal: Every 2 to 3 days

Notes

a) Upon initial thawing, a higher number of dead cells is observed, which is a normal phenomenon. Significant improvement is seen after adaptation. Once the cells reach a stable state, the number of dead cells decreases after subculturing and the cell growth rate becomes stable.

Related Products

CD28	
H_CD28 Reporter Jurkat Cell Line	Cynomolgus_CD28 CHO-K1 Cell Line
H_CD28 CHO-K1 Cell Line	H_CD28 HEK-293 Cell Line
Anti-CD28 hIgG4 Antibody(FR104)	Anti-H_CD28 hIgG4 Antibody(Theralizumab)
CD3	
Jurkat CD3-BsAb Reporter Cell Line	Cynomolgus_CD3E(Membrane Bound ECD) CHO-K1 Cell Line
H_CD3 CHO-K1 Cell Line	H_CD3 HEK-293 Cell Line
H_CD3E(Membrane Bound ECD) CHO-K1 Cell Line	Mouse_CD3 HEK-293 Cell Line
Anti-CD3 epsilon hIgG1 Antibody [OKT-3 (muromonab)]	Anti-CD3 hIgG1 Antibody(CH2527)



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