CHO-K1 human B2M FCGRT Cell Line

Cat. No: KC-1317 Version 19100402

Ⅰ. Cell Line Information

Catalog number	KC-1317
Cell line name:	CHO-K1 human B2M FCGRT Cell Line
Gene ID/Accession #:	NM_004048 & NM_001136019.3
Host cell line	CHO-K1
Cell type:	Chinese hamster ovary cell line
Description:	CHOK1 cell line stable expressing exogenous human B2M gene and FCGRT gene
Quantity:	One vial of frozen cells ($5X10^6$ per vial)
Stability:	Stable in culture over a minimum of 10 passages
Application:	Drug screening and biological assays
Freeze medium:	70% F12K + 20% FBS + 10% DMSO
Propagation medium:	F12K + 10% FBS + 6ug/ml Puromycin
Selection marker:	Puromycin
Morphology:	Fibroblastoid cells growing as monolayer
Subculture:	Split saturated culture 1:4~1:8 every 2~3 days; seed out at about 1-2 x 10^5 cells/ml
Incubation:	37 °C with 5% CO_2
Doubling time:	Approximately 24 hours
Mycoplasma status:	Negative
Biosafety level:	1
Storage:	Liquid nitrogen immediately upon receiving

Ⅱ. Background

FCGRT (Fc Fragment of IgG Receptor And Transporter) encodes a receptor that binds the Fc region of monomeric immunoglobulin G. The encoded protein transfers immunoglobulin G antibodies from mother to fetus across the placenta. This protein also binds immunoglobulin G to protect the antibody from degradation. Alternative splicing results in multiple transcript variants. Diseases associated with FCGRT include Selective Igg Deficiency Disease and Immunodeficiency 43. Gene Ontology (GO) annotations related to this gene include antigen binding and beta-2-microglobulin binding. An important paralog of this gene is HLA-C. [provided by RefSeq, Apr 2009; provided by GeneCards Summary]

β 2 microglobulin also known as B2M is a component of MHC class I molecules, MHC class I molecules have α1, α2, and α3 proteins which are present on all nucleated cells (excludes red blood cells). β 2 microglobulin associates not only with the alpha chain of MHC class I molecules, but also with class I-like molecules such as CD1 and Qa. The protein has a predominantly beta-pleated sheet structure that can form amyloid fibrils in some pathological conditions. The encoded antimicrobial protein displays antibacterial activity in amniotic fluid. A mutation in this gene has been shown to result in hypercatabolic hypoproteinemia.

III. Cell Line Generation

CHOK1 human B2M FCGRT cell line was generated using lentiviral vector expressing human B2M FCGRT sequence.

IV. Characterization using FACS

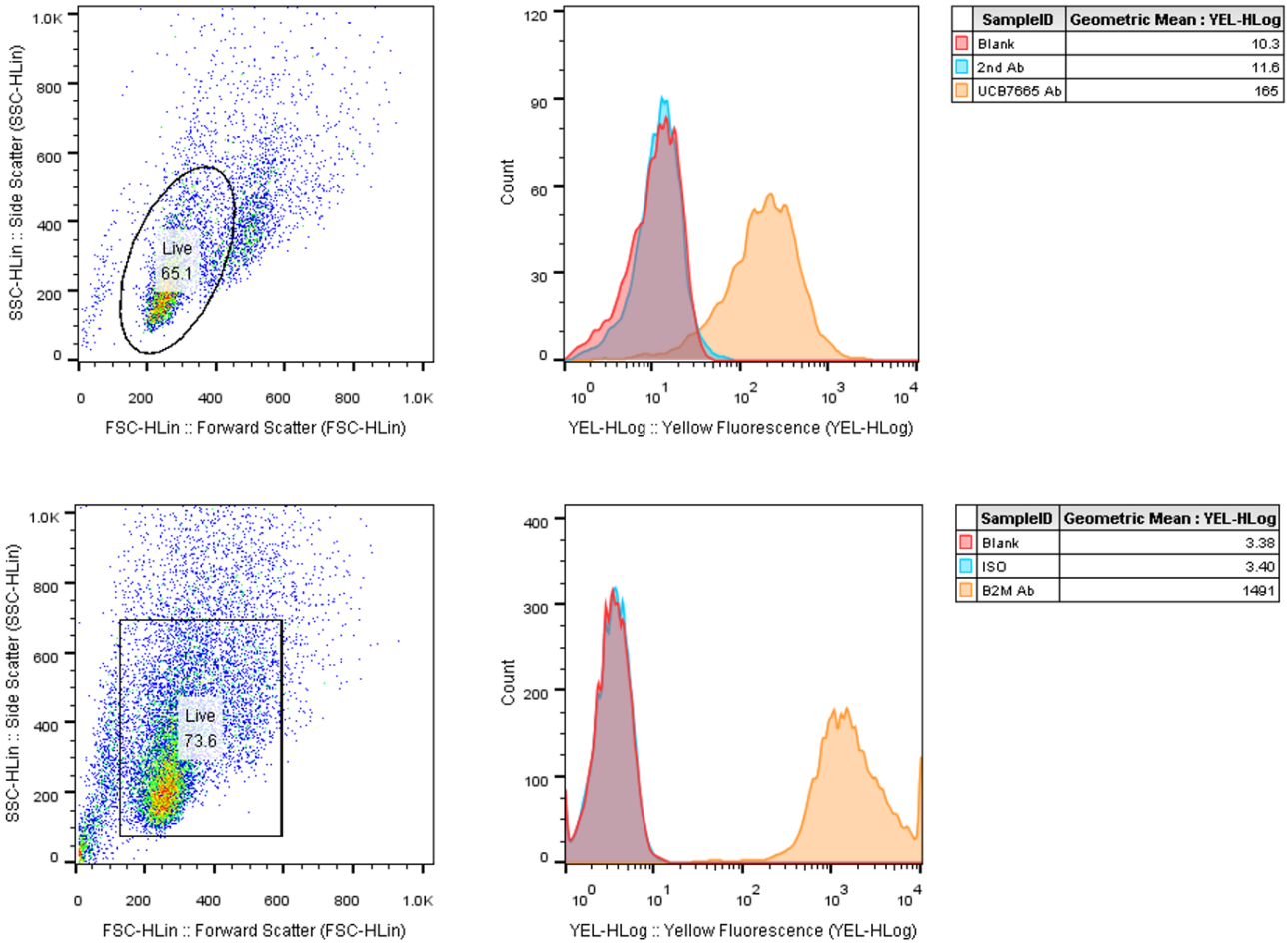

Figure: Characterization of B2M FCGRT overexpressing in CHOK1 stable clones using FACS.

V. Application

Hybridoma or Binders of ligand screening with FACS.

VI. Cell Resuscitation

1. Prewarm culture medium (F12K supplemented with 10% FBS and 6ug/ml puromycin) in a 37°C water bath.
2. Thaw the frozen vial in a 37°C water bath for 1-2 minutes.
3. Transfer the vial into biosafety cabinet and wipe the surface with 70% ethanol.
4. Unscrew the top of the vial and transfer the cell suspension gently into a sterile centrifuge tube containing 9.0 mL complete culture medium.
5. Spin at ~ 125 x g for 5~7 minutes at room temperature and discard the supernatant without disturbing the pellet.
6. Resuspend cell pellet with the appropriate volume of complete medium and transfer the cell suspension into a T25 culture flask.
7. Incubate the flask at 37°C, 5% CO_2 incubator.
8. Split saturated culture 1:4 ~ 1:8 every 2~3 days; seed out at about 1-2 x 10^5 cells/ml.

VII. Cell Freezing

1. Prepare the freezing medium (70% F12K + 20% FBS + 10% DMSO) fresh immediately before use.
2. Keep the freezing medium on ice and label cryovials.
3. Trypsin and harvest cells to a sterile, conical centrifuge tube during the logarithmic growth period and count the cells.
4. Centrifuge the cells at 250 x g for 5 minutes at room temperature and carefully aspirate off the medium.
5. Resuspend the cells at a density of at least 3 x10^6 cells/ml in chilled freezing medium.
6. Aliquot 1 ml of the cell suspension into each cryovial.
7. Freeze cells in the CoolCell freezing container overnight in a −80°C freezer.
8. Transfer vials to liquid nitrogen for long-term storage.

VIII. References

1. https://www.genecards.org/cgi-bin/carddisp.pl?gene=FCGRT
2. Stephen A. Beers. Influence of immunoglobulin isotype on therapeutic antibody function. Blood Spotlight. 2016
3. Güssow D, Rein R, Ginjaar I, Hochstenbach F, Seemann G, Kottman A, Ploegh HL (1 November 1987). "The human beta 2-microglobulin gene. Primary structure and definition of the transcriptional unit". J. Immunol. 139 (9): 3132–8. PMID 3312414

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