293T human CD122 Cell Line

Cat. No: KC-1007 Version 19051302

Ⅰ. Cell Line Information

Catalog number	KC-1007
Cell line name:	293T human CD122 Cell Line
Gene ID/Accession #:	NM_000878.5
Host cell line	293T
Cell type:	Human embryonic kidney
Description:	HEK293T cell line stable expressing exogenous human CD122 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% DMEM + 20% FBS + 10% DMSO
Propagation medium:	DMEM + 10% FBS + 0.5ug/ml Puromycin
Selection marker:	Puromycin
Morphology:	Fibroblastoid cells growing as monolayer
Subculture:	Split saturated culture 1:4~1:5 every 2~3 days; seed out at about 1-3 x 10^5 cells/ml
Incubation:	37 °C with 5% CO_2
Doubling time:	Approximately 30 hours
Mycoplasma status:	Negative
Biosafety level:	1
Storage:	Liquid nitrogen immediately upon receiving

Ⅱ. Background

CD122, also named as Interleukin-2 receptor subunit beta (IL2RB), which is involved in T cell-mediated immune responses, is present in 3 forms with respect to ability to bind interleukin 2. The low affinity form is a monomer of the alpha subunit (also called CD25) and is not involved in signal transduction. The intermediate affinity form consists of a gamma/beta subunit heterodimer, while the high affinity form consists of an alpha/beta/gamma subunit heterotrimer. Both the intermediate and high affinity forms of the receptor are involved in receptor-mediated endocytosis and transduction of mitogenic signals from interleukin 2. The protein encoded by this gene represents the beta subunit and is a type I membrane protein. This protein also forms one of the three subunits of

the IL-15 receptor. Activation of the receptor increases proliferation of CD8+ effector T cells

III. Cell Line Generation

293T human CD122 cell line was generated using lentiviral vector expressing human CD122 sequence.

IV. Characterization using FACS

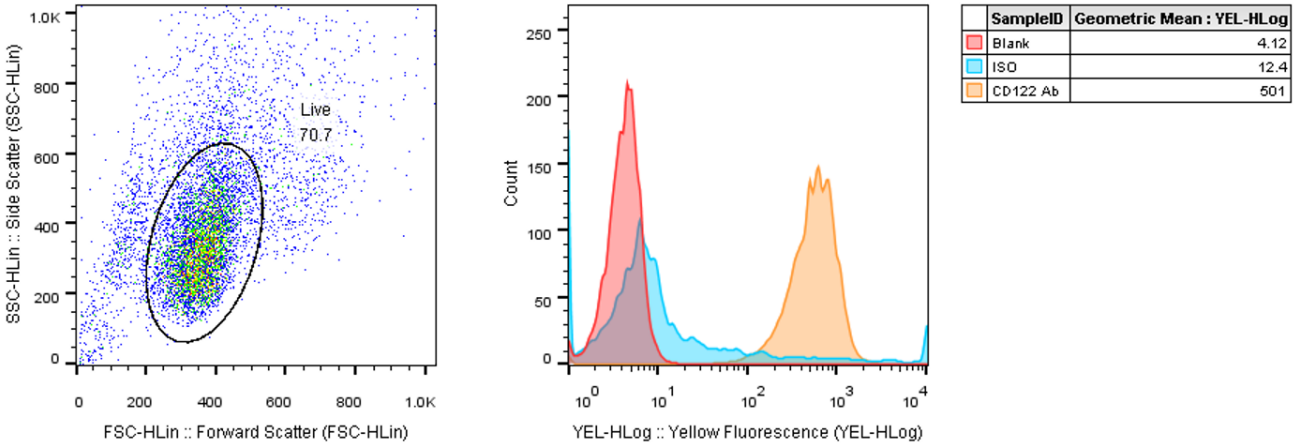

Figure: Characterization of CD122 overexpressing in 293T stable clones using FACS.

V. Application

Hybridoma or Binders of ligand screening with FACS.

VI. Cell Resuscitation

1. Prewarm culture medium (DMEM supplemented with 10% FBS and 0.5ug/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 ~ 1:5 every 2~3 days; seed out at about 1-3 x 10^5 cells/ml.

VII. Cell Freezing

1. Prepare the freezing medium (70% DMEM + 20% FBS + 10% DMSO) fresh immediately before use.
2. Keep the freezing medium on ice and label cryovials.
3. 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. Boyman O, Sprent J (February 17, 2012). "The role of interleukin-2 during homeostasis and activation of the immune system". Nat Rev Immunol. 12 (3): 180–190.
2. Miyazaki T, Kawahara A, Fujii H, Nakagawa Y, Minami Y, Liu ZJ, Oishi I, Silvennoinen O, Witthuhn BA, Ihle JN (November 1994). "Functional activation of Jak1 and Jak3 by selective association with IL-2 receptor subunits". Science. 266 (5187): 1045–7.

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