Human Recombinant KIR Stable Cell Line
Cat. No. M00650 **Version 08292017**

I. INTRODUCTION

Catalog Number: M00650
Cell Line Name: CHO-K1/KIR
Gene Synonyms: NKAT; NKAT1; p58.1; CD158A; KIR221; NKAT-1; KIR-K64
Expressed Gene: Codon Optimized from NM_014218.2; no expressed tags
Host Cell: CHO-K1
Quantity: Two vials of frozen cells (1×10^6 per vial)
Stability: 15 passages
Application: Binding assay or use as immunogen
Freeze Medium: 95% complete growth medium, 5% DMSO
Complete Growth Medium: F12K, 10% FBS
Culture Medium: F12K, 10% FBS, 8 μg/ml Puromycin
Mycoplasma Status[§]: Negative
Storage: Liquid nitrogen immediately upon receipt

II. BACKGROUND

Killer cell immunoglobulin-like receptors (KIRs) are transmembrane glycoproteins expressed by natural killer cells and are subsets of T cells. The KIR proteins are classified by the number of extracellular immunoglobulin domains (2D or 3D) and by whether they have a long (L) or short (S) cytoplasmic domain. KIR proteins with the long cytoplasmic domain transduce inhibitory signals upon ligand binding via an immune tyrosine-based inhibitory motif (ITIM), while KIR proteins with the short cytoplasmic domain lack the ITIM motif and instead associate with the TYRO protein tyrosine kinase binding protein to transduce activating signals. The ligands for several KIR proteins are subsets of HLA class I molecules; thus, KIR proteins are thought to play an important role in regulation of the immune response.

§: GenScript employs a PCR-based method to test the mycoplasma. The test covers 11 of the most common strains of mycoplasma, (covering approximately 95% of M. fermentans, M. hyorhinis, M. arginini, M. orale, M. salivarium, M. hominis, M. pulmonis, M. arthritidis, M. neurolyticum, M. hyopneumoniae and M. capricolum) and one species Ureaplasma (U. urealyticum), with sufficient sensitivity and specificity.

III. REPRESENTATIVE DATA

Protein Expression Validation

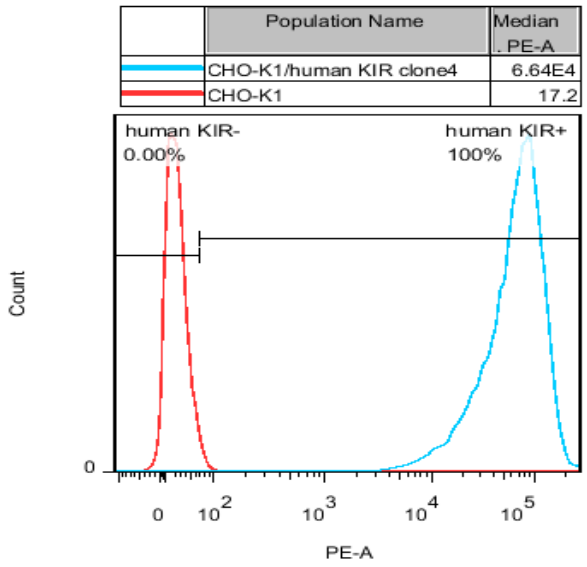

Figure 1. FACS analysis of KIR expression in CHO-K1 cells.

IV. THAWING AND SUBCULTURING

Thawing Protocol

1. Remove the vial from liquid nitrogen tank and thaw cells quickly in a 37°C water-bath.
2. Just before the cells are completely thawed, decontaminate the outside of the vial with 70% ethanol and transfer the cells to a 15 ml centrifuge tube containing 9 ml of complete growth medium.
3. Pellet cells by centrifugation at 200 x g for 5 minutes and remove the medium.
4. Re-suspend the cells in complete growth medium.
5. Transfer the cell suspension to a 10 cm dish with 10 ml of complete growth medium.
6. Grow the cells in an incubator at 37°C, 5 % CO_2.
7. Add antibiotic the following day.

Sub-culturing Protocol

1. Remove the culture medium from cells.
2. Wash cells with PBS (pH=7.4) to remove all traces of serum that contains trypsin inhibitor.
3. Add 2.0 ml of 0.25% (w/v) Trypsin- EDTA (GIBCO, Cat No. 25200-072) solution into 10 cm dish and observe the cells under an inverted microscope until cell layer is dispersed (usually within 3 to 5 minutes).
   Note: To avoid cells clumping, do not agitate the cells by hitting or shaking the dish while waiting for the cells to detach. If cells are difficult to detach, please place the dish in 37°C incubator for ~2 min.
4. Add 6.0 to 8.0 ml of complete growth medium into dish and aspirate cells by gently pipetting.
5. Centrifuge the cells at 200 x g for 5 minutes and remove the medium.
6. Re-suspend the cells in culture medium and add the cell suspension to a new culture dish.

7. Grow the cells in an incubator at 37°C, 5% CO_2.

Subcultivation Ratio: 1:4 to 1:8 weekly.
Medium Renewal: Every 2 to 3 days

V. REFERENCES

1. RefSeq. KIR2DL1 killer cell immunoglobulin-like receptor, two domains, long cytoplasmic tail [OL]. The National Center for Biotechnology Information, 2008.

GenScript USA Inc,
860 Centennial Ave.
Piscataway, NJ 08854
Toll-Free: 1-877-436-7274
Tel: 1-732-885-9188, Fax: 1-732-210-0262
Email: product@genscript.com
Web: http://www.genscript.com
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