Cyno Recombinant CD47 Stable Cell Line
Cat. No. M00632 **Version 06062017**

I. INTRODUCTION

Catalog Number: M00632
Cell Line Name: CHO-K1/cyno CD47
Gene Synonyms: N/A
Expressed Gene: Codon Optimized from NM_001266517; 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

This gene encodes a membrane protein, which is involved in the increase in intracellular calcium concentration that occurs upon cell adhesion to extracellular matrix. The encoded protein is also a receptor for the C-terminal cell binding domain of thrombospondin, and it may play a role in membrane transport and signal transduction. This gene has broad tissue, distribution, and is reduced in expression on Rh erythrocytes. Alternatively spliced transcript variants have been found for this gene.

§: 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

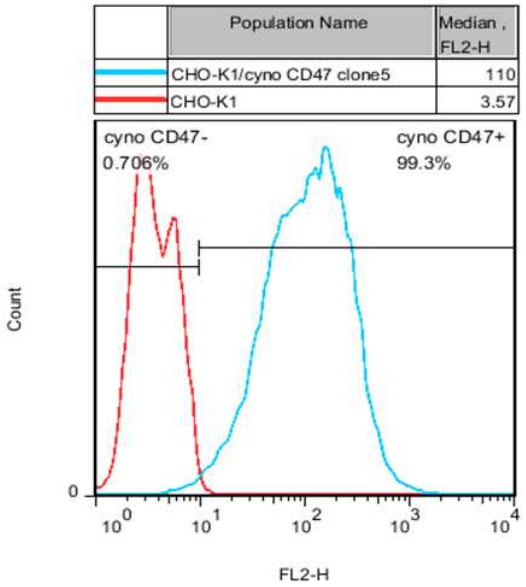

Figure 1. FACS analysis of cyno CD47 expression in CHO-K1 cells.

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 min, and remove the medium.
4. Resuspend 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 incubator with 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 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 min, and remove the medium.
6. Resuspend the cells in culture medium and add the cells suspension to new culture dish.
7. Grow the cells in incubator with 37°C, 5% CO_2.

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

V. REFERENCES

1.Yoshida K1, Tsujimoto H1, Matsumura K2, Kinoshita M3, Takahata R1, Matsumoto Y1, Hiraki S1, Ono S4, Seki S3, Yamamoto J1, Hase K1. CD47 is an adverse prognostic factor and a therapeutic target in gastric cancer [J]. Cancer Med, 2015 Sep; 4(9):1322-33.

2. Galli S1, Zlobec I2, Schürch C3, Perren A2, Ochsenbein AF4, Banz Y5. CD47 protein expression in acute myeloid leukemia: A tissue microarray-based analysis [J]. Leuk Res, 2015 Jul; 39(7):749-56.

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