Human Recombinant B7-H2 Stable Cell Line
Cat. No. M00671   **Version 08292017**

I.   INTRODUCTION

Catalog Number: M00671

Cell Line Name: CHO-K1/B7-H2

Gene Synonyms: B7H2; GL50; B7-H2; B7RP1; CD275; ICOSL; LICOS; B7RP-1; ICOS-L

Expressed Gene: Codon Optimized from NM_015259; 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

Ligand for the T-cell-specific cell surface receptor ICOS acts as a costimulatory signal for T-cell proliferation and cytokine secretion. It also induces B-cell proliferation and differentiation into plasma cells. It could also play an important role in mediating local tissue responses to inflammatory conditions, as well as modulating the secondary immune response by co-stimulating memory T-cell function.

III. REPRESENTATIVE DATA

Protein Expression Validation

Population Name	Median, FL4-H
CHO-K1/human B7-H2 clone3	1866
CHO-K1	1.49

human B7-H2 -
0.006%

human B7-H2 +
100.0%
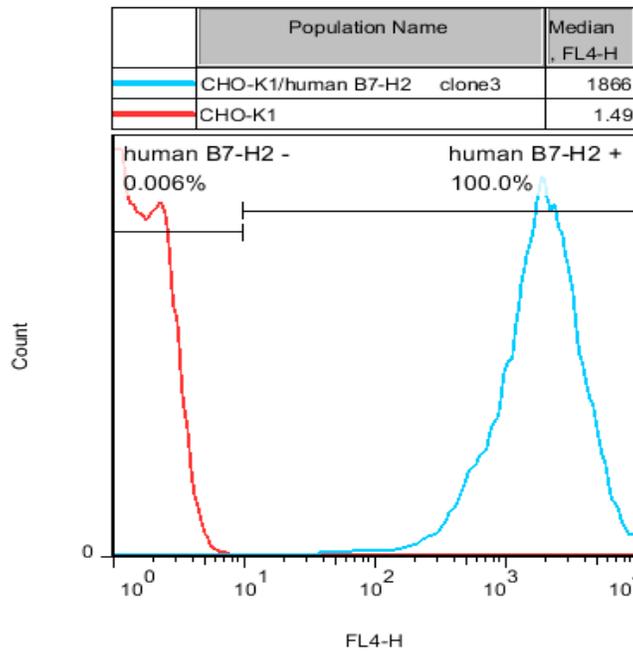
Figure 1.   FACS analysis of B7-H2 expression in CHO-K1 cells.

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 a 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 cell 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. Shengdian Wang, et.al. Costimulation of T cells by B7-H2, a B7-like molecule that binds ICOS [J]. Blood, 2000, 96: 2808-2813.

2. Faget J1, et.al. ICOS-ligand expression on plasmacytoid dendritic cells supports breast cancer progression by promoting the accumulation of immunosuppressive CD4+ T cells [J]. Cancer Res., 2012, 72(23): 6130-6141.

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