

Human Recombinant CD40 Stable Cell Line
Cat. No. M00606

Version 04282015

I. INTRODUCTION

Catalog Number: M00606

Cell Line Name: GS-H2/CD40

Gene Synonyms: Bp50, CDW40, TNFRSF5, p50

Expressed Gene: Codon Optimized from NM_001250.4; no expressed tags

Host Cell: GS-H2

Quantity: Two vials of frozen cells (1×10^6 per vial)

Stability: 20 passages

Application: *in vitro* functional assay

Freeze Medium: 95% complete growth medium, 5% DMSO

Complete Growth Medium: MEM, 10% FBS

Culture Medium: MEM, 10% FBS, 2 $\mu\text{g/ml}$ Puromycin, 200 $\mu\text{g/ml}$ Hygromycin B

Mycoplasma Status: Negative

Functional Performance: For CD40L protein, Signal / Background (S/B) > 3

Storage: Liquid nitrogen immediately upon receipt

II. BACKGROUND

Cluster of differentiation 40, CD40 is a costimulatory protein found on antigen presenting cells and is required for their activation. The binding of CD154 (CD40L) on TH cells to CD40 activates antigen presenting cells and induces a variety of downstream effects.

III. REPRESENTATIVE DATA

- Protein Expression Validation

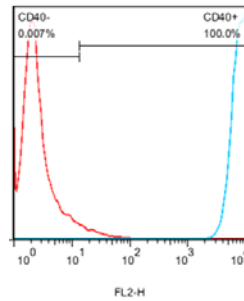


Figure 1. Flow cytometry analysis of CD40 expression in GS-H2/CD40 cells. Red: GS-H2, Blue: GS-H2/CD40.

- Validation by *in vitro* Functional Assay

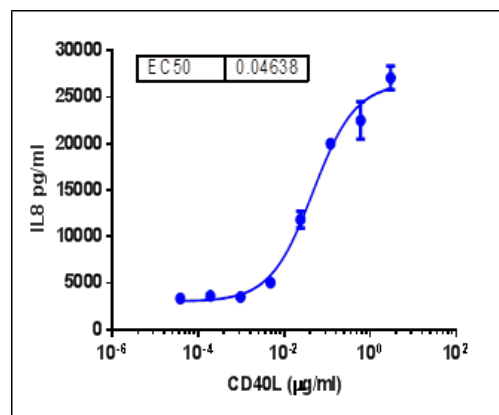


Figure 2. Functional analysis of CD40L protein on GS-H2/CD40 cells

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 % CO₂.
7. Add antibiotic in the following day.

Sub-culturing Protocol

1. Centrifuge the cells at 200 x g for 5min, and remove the medium.
2. Resuspend the cells in culture medium and add the cells suspension to new culture dish.
3. Grow the cells in incubator with 37°C, 5 % CO₂.

Subcultivation Ratio: A subcultivation ratio of 1:4 to 1:8 is recommended

Medium Renewal: Every 2 to 3 days

V. REFERENCES

1. Chatzigeorgiou A, Lyberi M, Chatzilymperis G, Nezos A, Kamper E. CD40/CD40L signaling and its implication in health and disease. *BioFactors* (Oxford, England). 2009, 35(6): 474–83.
2. Raul Elgueta, Micah J. Benson, Victor C. de Vries, Anna Wasiuk, Yanxia Guo, and Randolph J. Noelle. Molecular mechanism and function of CD40/CD40L engagement in the immune system. *Immunol Rev.* 2009 May: 229(1).

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