

Human Recombinant CD40 Stable Cell Line

Cat. No.: FCC-136

■ Introduction

Cell Line Name:	CHO-K1/CD40
Gene Synonyms:	p50; Bp50; CDW40; TNFRSF5
Expressed Gene:	CD40
Host Cell:	CHO-K1, (ATCC® CCL-61™)
Quantity:	Two vials of frozen cells (>1×10 ⁶ cells/vial)
Application:	Binding assay or use as immunogen
Cryopreservation Medium:	70% Complete growth medium, 20% FBS, 10% (V/V) DMSO
Complete Growth Medium:	F12K, 10% FBS
Drug Screening:	12 µg/ml Puromycin
Mycoplasma Contamination:	Not detected
Storage:	Liquid nitrogen immediately upon receipt

■ Background

This gene is a member of the TNF-receptor superfamily. The encoded protein is a receptor on antigen-presenting cells of the immune system and is essential for mediating a broad variety of immune and inflammatory responses including T cell-dependent immunoglobulin class switching, memory B cell development, and germinal center formation. AT-hook transcription factor AKNA is reported to coordinately regulate the expression of this receptor and its ligand, which may be important for homotypic cell interactions. Adaptor protein TNFR2 interacts with this receptor and serves as a mediator of the signal transduction. The interaction of this receptor and its ligand is found to be necessary for amyloid-beta-induced microglial activation, and thus is thought to be an early event in Alzheimer disease pathogenesis. Mutations affecting

this gene are the cause of autosomal recessive hyper-IgM immunodeficiency type 3 (HIGM3). Multiple alternatively spliced transcript variants of this gene encoding distinct isoforms have been reported.

■ **Representative Data**

Protein Expression Validation

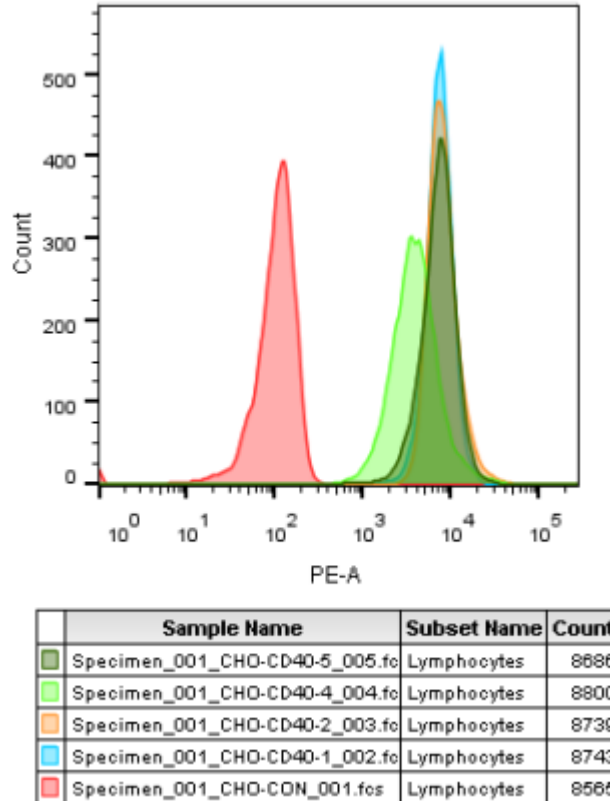


Figure 1: FACS analysis of CD40 expression in CHO-K1/CD40 cells

■ **Cell Culture Procedure**

1. Complete Growth Medium

The base medium for this cell line is ATCC-formulated F-12K Medium, Catalog No. 30-2004. To make the complete growth medium, add the following components to the base medium: fetal bovine serum to a final concentration of 10%.

2. Subculturing

- 1) Remove and discard culture medium.
- 2) Briefly rinse the cell layer with 0.25% (w/v) Trypsin- 0.53 mM EDTA solution to remove all traces of serum which contains trypsin inhibitor.

- 3) Add 2.0 mL of Trypsin-EDTA solution to flask and observe cells under an inverted microscope until cell layer is dispersed (usually within 2 to 5 minutes). **Note:** To avoid clumping do not agitate the cells by hitting or shaking the flask while waiting for the cells to detach. Cells that are difficult to detach may be placed at 37°C to facilitate dispersal.
- 4) Add 6.0 to 8.0 mL of complete growth medium and aspirate cells by gently pipetting.
- 5) Add appropriate aliquots of the cell suspension to new culture vessels.
- 6) Incubate cultures at 37°C.

Subcultivation Ratio: A subcultivation ratio of 1:3 to 1:5 is recommended.

Medium Renewal: Once or twice between subculture.

3. Cryopreservation

Freeze medium: 70% Complete growth medium, 20% FBS, 10% (V/V) DMSO

Storage temperature: liquid nitrogen vapor phase

4. Culture Conditions

Temperature: 37°C,

CO₂ : 5%

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