

Human Recombinant B7-H3 Stable Cell Line

Cat. No. M00536

Version 04282015

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I. INTRODUCTION

Catalog Number: M00536

Cell Line Name: CHO-K1/B7-H3

Gene Synonyms: CD276 ; 4Ig-B7-H3; B7H3; B7RP-2

Expressed Gene: Codon Optimized from NM_001024736.1; 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, 6 μ g/ml Puromycin

Mycoplasma Status: Negative

Storage: Liquid nitrogen immediately upon receipt

II. BACKGROUND

B7-H3, a new member of the B7 family of immune-regulatory molecules, was identified in 2001 by database searches of a human dendritic cell-derived cDNA library. It is a type I transmembrane protein, which expresses in certain normal cells and tissues, such as dendritic cells, as well as the liver, lung, breast, placenta, and prostate. Aberrant expression of B7-H3 has been reported in a wide range of solid cancers, including brain, lung, pancreatic, colorectal, liver, and breast cancers, as well as in hematologic malignancies, such as acute leukemia and multiple myeloma, and it is associated with more advanced disease and poor prognosis.

§: 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

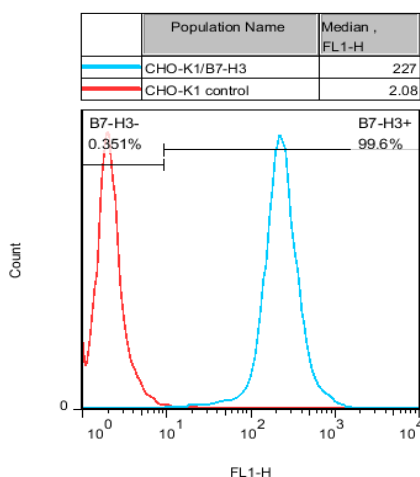


Figure 1. FACS analysis of B7-H3 expression in CHO-K1/B7-H3 cells.

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 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 0.05% (w/v) Trypsin- EDTA (GIBCO, Cat No. 25300) 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 5min, 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 %CO₂.

Subcultivation Ratio: 1:3 to 1:8 weekly.

Medium Renewal: Every 2 to 3 days

* GenScript uses a PCR-based method to test the mycoplasma. Our PCR mycoplasma test covers 11 of the most common species of mycoplasma (M. fermentans, M. hyorhinis, M. arginini, M. orale, M. salivarium, M. hominis, M. pulmonis, M. arthritidis, M. neurolyticum, M. hyopneumoniae, M. capricolum) and one species of Ureaplasma (U. urealyticum), with sufficient sensitivity and specificity.

V. REFERENCES

1. Mahoney KM1, Rennert PD2, Freeman GJ3. Combination cancer immunotherapy and new immunomodulatory targets. Nat Rev Drug Discov. 2015 Jul 31;14 (8):561-84.
2. Chapoval AI, Ni J, Lau JS et al. (2001). "B7-H3: a costimulatory molecule for T cell activation and IFN-gamma production". Nat. Immunol. 2 (3): 269–74.

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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