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<u>Data Sheet</u> BCMA—CHO Recombinant Cell Line (Low Expression) Catalog #79500-L

Description

Recombinant clonal stable CHO cell line constitutively expressing full length human BCMA protein, also known as TNFRSF17, (Genbank #NM_001192). Surface expression of BCMA was confirmed by flow cytometry. Each stable clonal cell line was selected for different levels of BCMA expression, (H. M, L), to mimic different stages of cancer target cells with various BCMA expression levels.

Background

B-cell maturation antigen (BCMA), also known as CD269 or tumor necrosis factor receptor superfamily member 17 (TNFRSF17), is a cell surface receptor of the TNF receptor superfamily that recognizes B-cell activating factor (BAFF). BCMA is preferentially expressed in mature B lymphocytes and also on Multiple Myeloma (MM) cells. BCMA is a highly attractive target antigen for immunotherapy because of its restricted expression in non-malignant tissue but almost universal expression on MM cells. CART-BCMA is an autologous T cell product engineered by lentiviral transduction to express a fully human BCMA-specific CAR (chimeric antigen receptor). CAR T cells targeting BCMA have pre-clinical anti-MM activity, and in 2017, the FDA granted BCMA CAR T-Cell immunotherapy breakthrough designation in Multiple Myeloma.

Application

- Useful as BCMA-expressing target cells in co-culture assay with BCMA-CAR-T cell, for both BCMA-specific cell killing assay and cytokine production assay.
- 2. Useful for screening and validating antibodies against BCMA and anti-BCMA CAR-T for immunotherapy research and drug discovery.
- 3. Useful for BCMA binding assays to screening for BCMA ligands.

Host Cell

CHO K1 cell line, Chinese Hamster Ovary

Format

Each vial contains ~ 2 x 10⁶ cells in 1 ml of 10% DMSO in FBS.

Storage

Store in liquid nitrogen immediately upon receipt.



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Cell Culture

Thaw Medium 3 (BPS Bioscience, #60186): Ham's F-12 medium (Hyclone #SH30526.01) supplemented with 10% FBS (Life technologies #26140-079), 1% Penicillin/Streptomycin (Hyclone #SV30010.01).

Growth Medium 3D (BPS Bioscience, #79539): Thaw Medium 3 (BPS Bioscience, #60186) plus 1 mg/ml G418 (Thermo Fisher, #11811031).

Recommended Culture Condition

Frozen Cells: Prepare a 50 ml conical tube with 10 ml of pre-warmed Thaw Medium 3 (**no G418**). Quickly thaw cells in a 37°C water bath with constant and slow agitation. Clean the outside of the vial with 70% ethanol and immediately transfer the entire content to Thaw Medium 3 (**no G418**). Avoid pipetting up and down, and gently rock the conical tube.

Spin the cells down at 150 x g for 5 minutes. Discard the medium and re-suspend the cell pellet in fresh Thaw Medium 3 (**no G418**). Transfer the entire content to a T25 flask to distribute the cells. Incubate the cells in a humidified 37° C incubator with 5% CO₂. After 48-72 hours of incubation, change to fresh Thaw Medium 3 (**no G418**), without disturbing the attached cells. Continue to change the medium every 2-3 days until the cells reach desired confluency. If slow cell growth occurs during resuscitation, increase FBS to 15% for the first week of culture. Switch to Growth Medium 3D after the first passage.

Subculture: When cells reach 90% confluency, remove the medium and GENTLY wash once with PBS (without Magnesium or Calcium). These cells are loosely adherent and detach easily so do not re-suspend the PBS directly onto the cell surface. Treat cells with 2 ml of 0.25% trypsin/EDTA and incubate for 2-3 minutes at 37°C. After confirming cell detachment by light microscopy, add 10 ml pre-warmed medium and gently pipette up and down to dissociate cell clumps. Transfer cells to a 15 ml conical tube and centrifuge at 200 x g for 5 minutes. Remove the medium and re-suspend cells in 10 ml of pre-warmed Growth Medium 3D. Dispense 5 ml of the cell suspension into a new T75 flask containing 20 ml pre-warmed media. Incubate cells in a humidified 37°C incubator with 5% CO₂. Freeze cells in freezing medium (10% DMSO in FBS) when cells reach 90% confluency. Cells have been demonstrated to be stable for at least 15 passages; BPS recommends preparing frozen stocks at an early passage so cells are not used beyond passage 20.

Mycoplasma Testing

This cell line has been screened using the MycoAlert™ Mycoplasma Detection Kit (Lonza, #LT07-118) to confirm the absence of Mycoplasma contamination. MycoAlert Assay Control Set (Lonza, #LT07-518) was used as a positive control.

Application References

- 1. Ghosh A, *et al.* CAR T cell therapy for multiple myeloma *Leuk Lymphoma. 2017* Nov; **6:**1-12.
- 2. Sanchez E, *et al.* The clinical significance of B-cell maturation antigen as a therapeutic target and biomarker.

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Expert Rev Mol Diagn. 2018 Mar; 7:1-11.

- 3. Sohail A., *et al.* Emerging immune targets for the treatment of multiple myeloma. *Immunotherapy.* 2018 Feb 1; **10(4):**265-282.
- 4. Sidaway P., et al. Anti-BCMA CAR T cells show promise in MM. Nat Rev Clin Oncol. 2016 Sep;13(9):530.

Vector and Sequence

Human BCMA (NM_001192) was cloned into pIRESneo.

MLQMAGQCSQNEYFDSLLHACIPCQLRCSSNTPPLTCQRYCNASVTNSVKGTNAILWTCLGLS LIISLAVFVLMFLLRKINSEPLKDEFKNTGSGLLGMANIDLEKSRTGDEIILPRGLEYTVEECTCEC IKSKPKVDSDHCFPLPAMEEGATILVTTKTNDYCKSLPAALSATEIEKSISAR

Quality Assurance

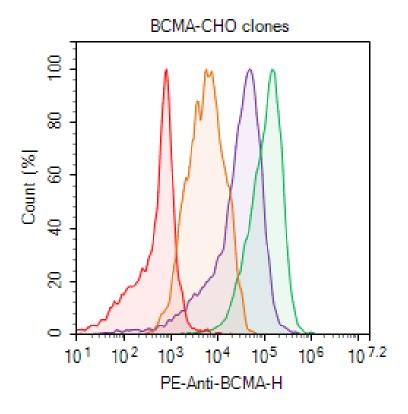


Figure 1. Expression of BCMA validated by flow cytometry. Flow cytometry using PEconjugated anti-human BCMA (CD269) antibody (Biolegend, #357504) detects BCMA surface expression of BCMA-CHO Recombinant Cell Lines with different expression levels: #79500-H, high expresser: green; #79500-M, medium expresser: purple; #79500-L, low expresser: brown; WT CHO negative control: red.

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Related Products

Product	Cat. #	Size
BCMA— CHO Recombinant Cell Line (High Expression)	79500-H	2 vials
BCMA— CHO Recombinant Cell Line (Medium Expression)	79500-M	2 vials
Thaw Medium 3	60186	100 ml
Growth Medium 3D	79539	500 ml
CD22 CHO Recombinant Cell Line (High Expression)	79557-H	2 vials
CD22 CHO Recombinant Cell Line (Medium Expression)	79557-M	2 vials
CD19 CHO Recombinant Cell Line (High Expression)	79561-H	2 vials
CD19 CHO Recombinant Cell Line (Medium Expression)	79561-M	2 vials
CD19 CHO Recombinant Cell Line (Low Expression)	79561-L	2 vials
Human BCMA (CD269)	90105-A	5 µg
Human BCMA (CD269)	90105-B	20 µg
Human BCMA, Fc-Fusion, Avi-Tag HiP™	79465	100 µg
Human BCMA, Fc-fusion (IgG1), Avi-Tag, Biotin-Labeled HiP™	79467	50 µg
Human BAFF	90100-1	10 µg
Human BAFF	90100-2	100 µg
Human BAFF	90100-3	1 mg
Human BAFF-R(CD268)	90103-A	10 µg
Human BAFF-R(CD268)	90103-B	50 µg