

Product Information

Description:	Anti-BCMA-Anti-CD3-Avi-His-Tag is a purified recombinant human bispecific molecule with T cell Engager. This bispecific molecule has been tested for specific activity in both ELISA binding assay to BCMA-biotin and functional reporter assay using NFAT-luc reporter Jurkat cell line (BPS Bioscience #60621) in the presence of BCMA-CHO cells (BPS Bioscience #79500-H).
Background:	B-cell maturation antigen (BCMA), also known as tumor necrosis factor receptor superfamily member 17 (TNFRSF17), is a protein encoded by the TNFRSF17 gene. 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. Upregulation of BCMA also correlates with disease burden and prognosis in multiple myeloma. This bispecific molecule binds to BCMA on cancer cells and CD3 on T cells simultaneously, thus bringing T lymphocytes closer to the cancer cells. The binding event potentiates unstimulated T cells and induces direct cytotoxicity against BCMA+ cancer cells.
Concentration:	0.88 mg/ml
Species:	Human
Formulated In:	8 mM phosphate pH 7.4, 110 mM NaCl, 2.2 mM KCl, and 20% glycerol
Expression System:	HEK293
Format:	Aqueous buffer solution
Stability:	At least 12 months at -80°C. Avoid freeze/thaw cycles.
Storage:	-80°C
MW:	57 kDa + glycans
Glycosylation:	This protein runs at a higher MW by SDS-PAGE due to glycosylation.
Purity:	≥90%
Purification:	Ni-NTA affinity purification of the His-tag protein from HEK293 cells
Applications:	This product is for research use only. It is not suitable for human diagnostic or therapeutic use. The anti-BCMA-anti-CD3-Avi-His-Tag can be used for studying BCMA+ cancer cell-mediated T cell activation, using either primary T cells or reporter cell lines such as NFAT-luc-Jurkat cells (BPS Bioscience #60621).

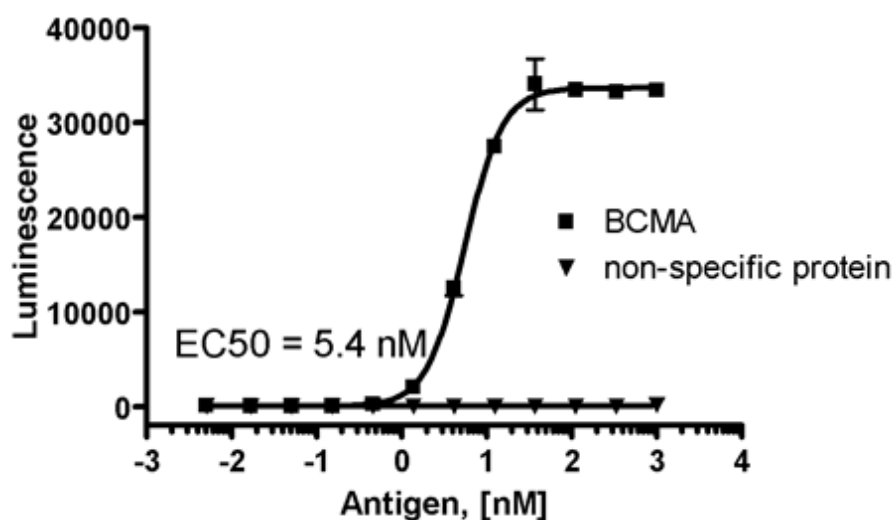
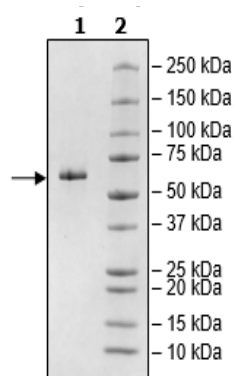
Experimental design and assay protocol for measuring anti BCMA-anti CD3 functional activity using NFAT-luc reporter Jurkat cell line:

1. Harvest BCMA CHO cells (BPS Bioscience #79500) from culture and seed cells at a density of 30,000 cells per well into a white clear-bottom 96-well microplate in 50 µl of assay medium (Thaw Medium 2 BPS Bioscience #60184). Incubate cells at 37° in a CO₂ incubator for several hours to allow them to attach.
2. Prepare a serial dilution of anti-BCMA/anti-CD3 molecule in assay medium in a fresh 96-well plate. The concentration of molecule here is 4x the final treatment concentration. Set up each treatment in at least triplicate. Add 25 ml of each dilution to the BCMA CHO cells.
3. Harvest the NFAT-reporter-Jurkat cells (BPS Bioscience #60621) by centrifugation and resuspend in assay medium. Dilute cells to 1.2 x 10⁶ / ml in assay medium. The final cell density of NFAT Reporter- Jurkat cells is 30,000 cells per well. Add 25 ml of NFAT-reporter-Jurkat cells to the BCMA CHO cells. The final volume will be 100 ml per well including both cell lines and the test molecule.
4. Add 100 µl of assay medium to cell-free control wells (for determining background luminescence). Incubate the plates at 37° in a CO₂ incubator overnight.

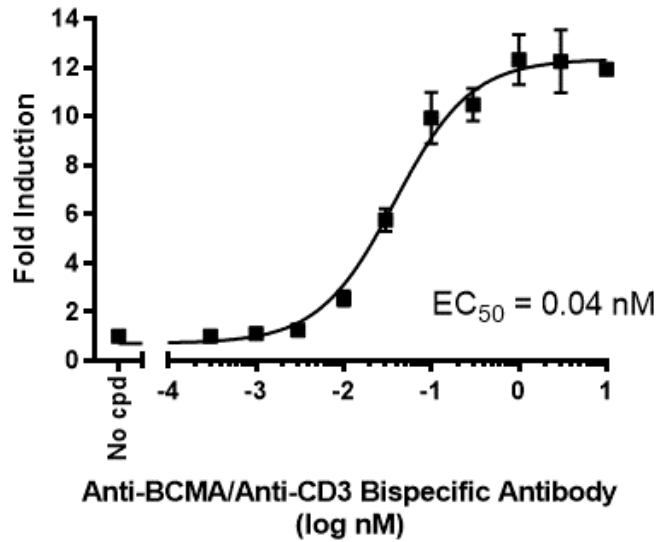
- The next day, perform the luciferase assay using the **ONE-Step™ Luciferase Assay System** (BPS Bioscience #60690). Add 100 μ l of **ONE-Step™** Luciferase reagent per well and rock gently at room temperature for ~15 minutes. Measure luminescence using a luminometer. *If using luciferase reagents from other vendors, follow the manufacturer's assay protocol.*
- Data Analysis: Subtract the average background luminescence (cell-free control wells) from the luminescence reading of all wells. The fold induction of NFAT luciferase reporter expression = background-subtracted luminescence of treated well / average background-subtracted luminescence of untreated control wells.

Quality Control Data

4-20% SDS-Page Coomassie Staining



ELISA assay shows specific binding of anti BCMA-anti CD3 when the ELISA plate is coated with anti BCMA-anti CD3 molecule and exposed to a BCMA Biotin (BPS Bioscience #79467-1) titration.



Activation of NFAT Reporter Jurkat cells by anti BCMA-anti CD3 bispecific molecule in the presence of BCMA-CHO cells.