Biotinylated Human B7-1 / CD80 Protein, Fc,Avitag™ (MALS verified)

Catalog # B71-H82F2



Synonym

CD80,B7,B7-1,B7.1,BB1,CD28LG,CD28LG1,LAB7

Source

Biotinylated Human B7-1, Fc,Avitag(B71-H82F2) is expressed from human 293 cells (HEK293). It contains AA Val 35 - Asn 242 (Accession # NP_005182.1). Predicted N-terminus: Val 35

Molecular Characterization

B7-1(Val 35 - Asn 242) Fc(Pro 100 - Lys 330)
NP_005182.1 P01857

This protein carries a human IgG1 Fc tag at the C-terminus, followed by an Avi tag (AvitagTM).

The protein has a calculated MW of 52.3 kDa. The protein migrates as 65-95 kDa under reducing (R) condition, and 130-160 kDa under non-reducing (NR) condition (SDS-PAGE) due to glycosylation.

Labeling

Biotinylation of this product is performed using AvitagTM technology. Briefly, the single lysine residue in the Avitag is enzymatically labeled with biotin.

Protein Ratio

Passed as determined by the HABA assay / binding ELISA.

Endotoxin

Less than 1.0 EU per µg by the LAL method.

Purity

>95% as determined by SDS-PAGE.

Formulation

Lyophilized from 0.22 µm filtered solution in

Tris with Glycine, Arginine and NaCl, pH7.5 with trehalose as protectant.

Contact us for customized product form or formulation.

Reconstitution

Please see Certificate of Analysis for specific instructions.

For best performance, we strongly recommend you to follow the reconstitution protocol provided in the CoA.

Storage

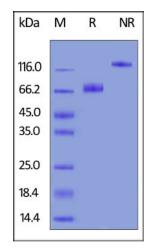
For long term storage, the product should be stored at lyophilized state at -20°C or lower.

Please avoid repeated freeze-thaw cycles.

This product is stable after storage at:

- -20°C to -70°C for 12 months in lyophilized state;
- -70°C for 3 months under sterile conditions after reconstitution.

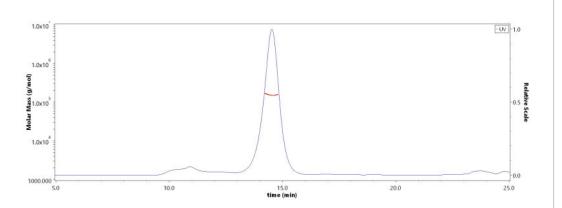
SDS-PAGE



Biotinylated Human B7-1, Fc, Avitag on SDS-PAGE under reducing (R) and non-reducing (NR) conditions. The gel was stained with Coomassie Blue. The purity of the protein is greater than 95%.

Bioactivity-ELISA

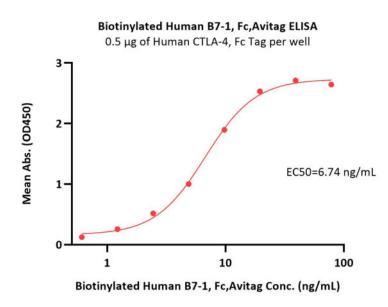
SEC-MALS



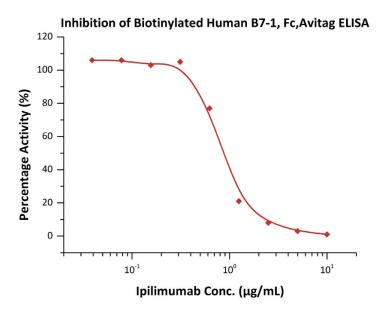
The purity of Biotinylated Human B7-1, Fc, Avitag (Cat. No. B71-H82F2) is more than 85% and the molecular weight of this protein is around 130-165 kDa verified by SEC-MALS.

Report



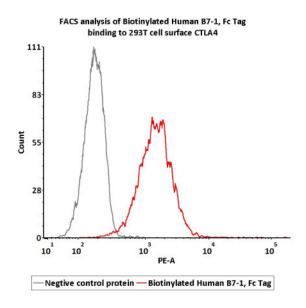


Immobilized Human CTLA-4, Fc Tag (Cat. No. CT4-H5255) at 5 μ g/mL (100 μ L/well) can bind Biotinylated Human B7-1, Fc,Avitag (Cat. No. B71-H82F2) with a linear range of 0.6-9.8 μ g/mL (QC tested).

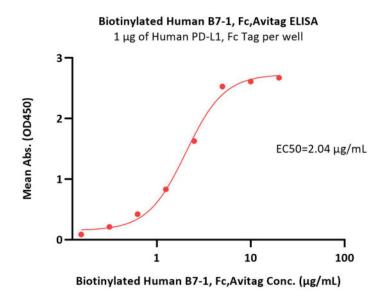


Serial dilutions of Ipilimumab were added into Human CTLA-4, Fc Tag (Cat. No. CT4-H5255): Biotinylated Human B7-1, Fc,Avitag (Cat. No. B71-H82F2) binding reactions. The half maximal inhibitory concentration (IC50) is $0.8260 \, \mu \text{g/mL}$ (Routinely tested).

Bioactivity-FACS

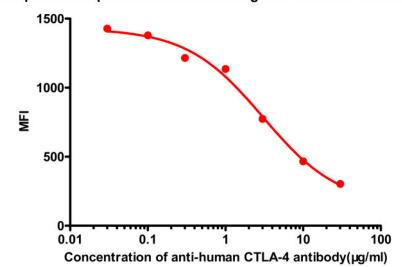


Flow Cytometry assay shows that Biotinylated Human B7-1, Fc,Avitag (Cat. No. B71-H82F2) can bind to 293 cell overexpressing human CTLA-4. The concentration of Human B7-1 is 0.3 μ g/mL (Routinely tested).



Immobilized Human PD-L1, Fc Tag (Cat. No. PD1-H5258) at $10 \mu g/mL$ (100 $\mu L/well$) can bind Biotinylated Human B7-1, Fc,Avitag (Cat. No. B71-H82F2) with a linear range of 0.156-5 $\mu g/mL$ (Routinely tested).

Competitive experiment of neutralizing anti-human CTLA-4 antibody



FACS analysis shows that the binding of Biotinylated Human B7-1, Fc, Avitag (Cat. No. B71-H82F2) to 293 overexpressing CTLA-4 was inhibited by increasing concentration of neutralizing Anti-Human CTLA-4 antibody. The



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concentration of B7-1 used is 0.3 μ g/mL. The IC50 is 3.025 μ g/mL (Routinely tested).

Background

B7-1 and B7-2, together with their receptors CD28 and CTLA4, constitute one of the dominant co-stimulatory pathways that regulate T and Bcell responses. Although both CTLA4 and CD28 can bind to the same ligands, CTLA4 binds to B71 and B72 with a 20 100 fold higher affinity than CD28 and is involved in the downregulation of the immune response.

B-lymphocyte activation antigen B7-1 (referred to as B7) also known as cluster of Differentiation 80 (CD80), is a member of cell surface immunoglobulin superfamily and is expressed on activated B cells, activated T cells, macrophages and dendritic cells. It is the ligand for two different proteins on the T cell surface: CD28 (for autoregulation and intercellular association) and CTLA-4 (for attenuation of regulation and cellular disassociation). CD80 works in tandem with CD86 to prime T cells. CD80 plays a role in induction of innate immune responses by activating NF-κB-signaling pathway in macrophages. CD80 is thus regarded as promising therapeutic targets for autoimmune diseases and various carcinomas.

Clinical and Translational Updates

