FITC-Labeled Human CD20 / MS4A1 Full Length Protein, His Tag (Nanodisc, SPR verified)

Catalog # CD0-HF2H5



Synonym

MS4A1,CD20,MS4A-1

Source

FITC-Labeled Human CD20 Full Length Protein, His Tag(CD0-HF2H5) is expressed from human 293 cells (HEK293). It contains AA Met 1 - Pro 297 (Accession # P11836-1).

Predicted N-terminus: Met 1

Molecular Characterization

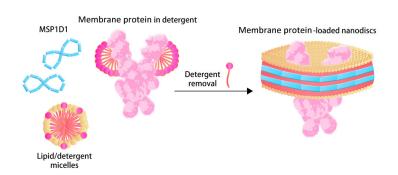
CD20(Met 1 - Pro 297) P11836-1

Poly-his

This protein carries a polyhistidine tag at the C-terminus.

The protein has a calculated MW of 35.2 kDa. The protein migrates as 26 kDa, 40 kDa and 70 kDa when calibrated against <u>Star Ribbon Pre-stained Protein</u> <u>Marker</u> under reducing (R) condition (SDS-PAGE) due to glycosylation.

Nanodiscs are a new class of model membranes that are being used to solubilize and study a range of integral membrane proteins and membrane-associated proteins. The Nanodisc bilayer is bounded by a membrane scaffold protein (MSP1D1) coat that confers enhanced stability and a narrow particle size distribution.



The nanodisc assembles from a mixture of full length membrane protein in detergent, phospholipid micelles and membrane scaffold protein(MSP1D1) upon removal of the detergent.

Conjugate

FITC

Excitation source: 488 nm spectral line, argon-ion laser

Excitation Wavelength: 488 nm Emission Wavelength: 535 nm

Labeling

The primary amines in the side chains of lysine residues and the N-terminus of the protein are conjugated with FITC using standard chemical labeling method. The residual FITC is removed by molecular sieve treatment during purification process.

*The isotype control of empty/mock nanodisc (Cat. No. APO-H51H3) is sold separately and not included in protein, you can follow this link for product information.

Purity

>85% as determined by SDS-PAGE.

Formulation

Supplied as 0.2 μ m filtered solution in 50 mM HEPES, 150 mM NaCl, pH7.5 with trehalose as protectant.

Contact us for customized product form or formulation.

Shipping

This product is supplied and shipped with dry ice, please inquire the shipping cost.

Storage

Please protect from light and avoid repeated freeze-thaw cycles.

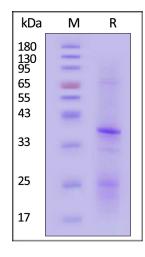
This product is stable after storage at:

- The product MUST be stored at -70°C or lower upon receipt;
- -70°C for 3 months under sterile conditions.



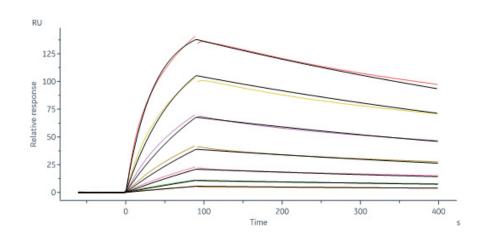


SDS-PAGE

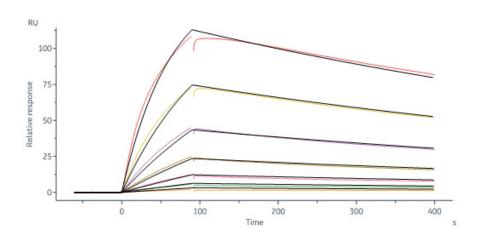


FITC-Labeled Human CD20 Full Length Protein, His Tag on SDS-PAGE under reducing (R) condition. The gel was stained with Coomassie Blue. The purity of the protein is greater than 85% (With <u>Star Ribbon Pre-stained Protein Marker</u>).

Bioactivity-SPR



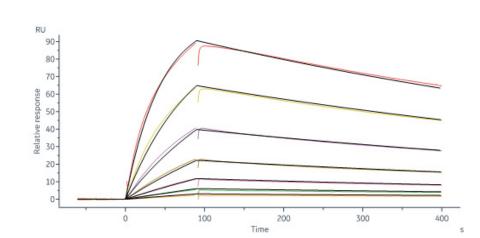
Obinutuzumab captured on Protein A Chip can bind FITC-Labeled Human CD20 Full Length Protein, His Tag (Cat. No. CD0-HF2H5) with an affinity constant of 23.4 nM as determined in a SPR assay (Biacore 8K) (QC tested).



Ofatumumab captured on Protein A Chip can bind FITC-Labeled Human CD20 Full Length Protein, His Tag (Cat. No. CD0-HF2H5) with an affinity constant of 37.3 nM as determined in a SPR assay (Biacore 8K) (Routinely tested).

Bioactivity-FACS



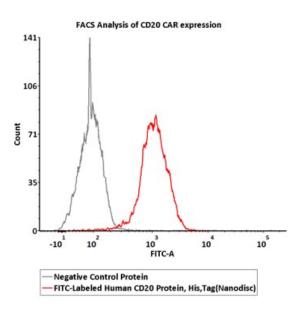


Rituximab captured on Protein A Chip can bind FITC-Labeled Human CD20 Full Length Protein, His Tag (Cat. No. CD0-HF2H5) with an affinity constant of 27.3 nM as determined in a SPR assay (Biacore 8K) (Routinely tested).

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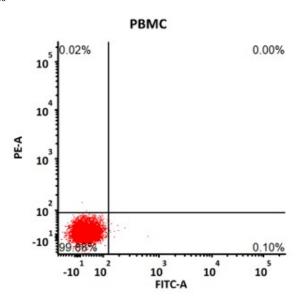


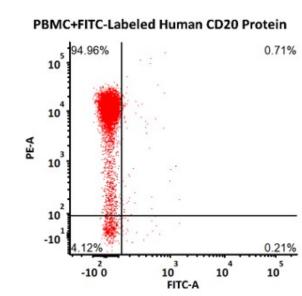


Flow cytometric analysis of Anti-CD20 CAR-293 cells staining with FITC-Labeled Human CD20 Full Length Protein, His Tag (Cat. No. CD0-HF2H5) at 1 μ g/mL (1 μ g/mL corresponds to labeling of 2.5e5 cells in a final volume of 100 μ L), compared with negative control protein. FITC signal was used to evaluate the binding activity (QC tested).

Evaluation of CAR expression

FACS Analysis of Non-specific binding to PBMCs





Non-specificity of FITC-Labeled Human CD20 Full Length Protein, His Tag (Cat. No. CD0-HF2H5) binding to CD3+ cells present in human PBMC. 5e5 of human PBMCs were simultaneously stained with PE-labeled anti-CD3 antibody and FITC-Labeled Human CD20 Full Length Protein, His Tag (1 µg/mL corresponds to labeling of 5e5 cells in a final volume of 100 µL) and washed and then analyzed with FACS. Both FITC and PE positive signals was used to evaluate the non-specific binding activity to human CD3+ cells (QC tested).

Background

B-lymphocyte antigen CD20 is also known as B-lymphocyte surface antigen B1, Leukocyte surface antigen Leu-16, Membrane-spanning 4-domains subfamily A member 1 and MS4A1, is an activated-glycosylated phosphoprotein expressed on the surface of all B-cells beginning at the pro-B phase (CD45R+, CD117+) and progressively increasing in concentration until maturity. CD20 is expressed on all stages of B cell development except the first and last; it is present from late pro-B cells through memory cells, but not on either early pro-B cells or plasma blasts and plasma cells. It is found on B-cell lymphomas, hairy cell leukemia, B-cell chronic lymphocytic leukemia, and melanoma cancer stem cells. The protein has no known natural ligand and its function is to enable optimal B-cell immune response, specifically against T-independent antigens. It is suspected that it acts as a calcium channel in the cell membrane. CD20 / MS4A1 is the target of the monoclonal antibodies (mAb) rituximab, Ibritumomab tiuxetan, and tositumomab, which are all active agents in the treatment of all B cell lymphomas and leukemias. Defects in CD20 / MS4A1 are the cause of immunodeficiency common variable type 5 (CVID5); also called antibody deficiency due to CD20 defect. CVID5 is a primary



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immunodeficiency characterized by antibody deficiency, hypogammaglobulinemia, recurrent bacterial infections and an inability to mount an antibody response to antigen.

