Product Datasheet

CD36 Antibody (1283D) - BSA Free NBP2-54790

Unit Size: 0.1 mg

Store at 4C short term. Aliquot and store at -20C long term. Avoid freeze-thaw cycles.

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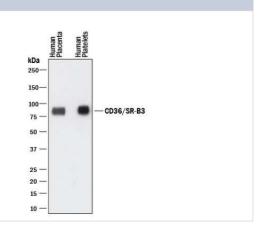
NBP2-54790

CD36 Antibody (1283D) - BSA Free

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Product Information	
Unit Size	0.1 mg
Concentration	1.0 mg/ml
Storage	Store at 4C short term. Aliquot and store at -20C long term. Avoid freeze-thaw cycles.
Clonality	Monoclonal
Clone	1283D
Preservative	0.02% Sodium Azide
Isotype	IgG
Purity	Protein A or G purified
Buffer	PBS
Target Molecular Weight	110 kDa
Product Description	
Description	Novus Biologicals Rabbit CD36 Antibody (1283D) - BSA Free (NBP2-54790) is a recombinant monoclonal antibody validated for use in IHC, WB, ICC/IF and IP. Anti-CD36 Antibody: Cited in 1 publication. All Novus Biologicals antibodies are covered by our 100% guarantee.
Host	Rabbit
Gene ID	948
Gene Symbol	CD36
Species	Human
Immunogen	This CD36 Antibody (1283D) was developed against a synthetic peptide made to an internal portion of the human CD36 protein (between amino acids 100-150) [Uniprot P16671]
Product Application Details	
Applications	Western Blot, Immunohistochemistry-Paraffin, Immunocytochemistry/ Immunofluorescence, Immunohistochemistry
Recommended Dilutions	Western Blot 1.0 ug/ml, Immunohistochemistry 5-10 ug/ml, Immunocytochemistry/ Immunofluorescence 10-20 ug/ml,

Images

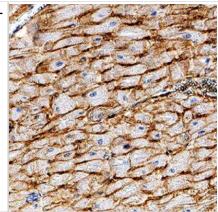
Western Blot: CD36 Antibody (1283D) [NBP2-54790] - Western blot shows lysates of human placenta tissue and human platelets. PVDF membrane was probed with 1 ug/ml of Rabbit Anti-Human CD36 Monoclonal antibody (catalog number NBP2-54790) followed by HRP-conjugated Anti-Rabbit IgG Secondary Antibody (catalog number HAF008). A specific band was detected for CD36/SR-B3 at approximately 85 kDa (as indicated). This experiment was conducted under reducing conditions.



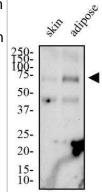


Immunohistochemistry-Paraffin 5-10 ug/ml

Immunohistochemistry-Paraffin: CD36 Antibody (1283D) [NBP2-54790] - CD36 was detected in immersion fixed paraffin-embedded sections of human heart using Rabbit Anti-Human CD36/SR-B3 Monoclonal Antibody for 1 hour at room temperature followed by incubation with the Anti-Rabbit IgG VisUCyte(TM) HRP Polymer Antibody. Tissue was stained using DAB (brown) and counterstained with hematoxylin (blue). Specific staining was localized to cardiomyocyte membranes.



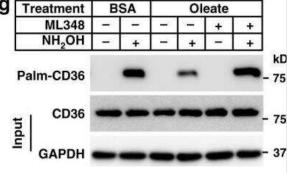
Western Blot: CD36 Antibody (1283D) [NBP2-54790] - Total protein from human skin and adipose tissue was separated on a 12% gel by SDS-PAGE, transferred to PVDF membrane and blocked in 5% non-fat milk in TBST. The membrane was probed with 1.0 ug/ml anti-CD36 in 1% non-fat milk in TBST and detected with an anti-rabbit HRP secondary antibody using chemiluminescence.



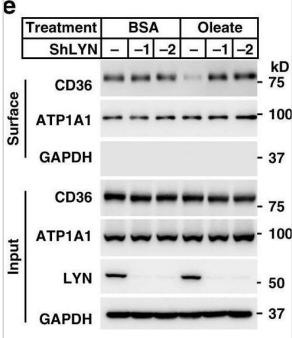
Immunocytochemistry/Immunofluorescence: CD36 Antibody (1283D) [NBP2-54790] - HepG2 cells were fixed for 10 minutes using 10% formalin and then permeabilized for 5 minutes using 1X TBS + 0.5% Triton-X100. The cells were incubated with anti-CD36 (1283D) at 5.0 ug/ml overnight at 4C and detected with an anti-rabbit Dylight 488 (Green) at a 1:500 dilution. Alpha tubulin (DM1A) NB100-690 was used as a co-stain at a 1:1000 dilution and detected with an anti-mouse Dylight 550 (Red) at a 1:500 dilution. Nuclei were counterstained with DAPI (Blue). Cells were imaged using a 40X objective..

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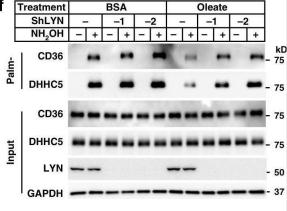
Western Blot: CD36 Antibody (1283D) - BSA Free [NBP2-54790] - 3T3-L1 adipocytes were pretreated and treated with ML348 (10 uM) for 1 h, followed by BSA or oleate treatment for another 1 h. Image collected and cropped by CiteAb from the following publication (//pubmed.ncbi.nlm.nih.gov/32958780/) licensed under a CC-BY license.



Western Blot: CD36 Antibody (1283D) - BSA Free [NBP2-54790] - LYN phosphorylates DHHC5 & is required for the endocytosis of FAs.a 3T3-L1 adipocytes pretreated with serum-free medium for 4 h & PP2 (15 µM) for 1 h, followed by treatment with BSA or oleate (100 µM) for 5 min. Cells harvested & immunoprecipitated with anti-DHHC5 antibody to detect phosphorylation of Tyr91. b 3T3-L1 adipocytes transduced with indicated shRNA set up & treated with oleate for 5 min. Cells harvested to detect Tyr91 phosphorylation as in (a). c On day 0, HEK293T cells set up at 2.5 × 105 cells per 6-cm dish. On day 2, cells transfected with 0.5 μg of WT, Y91E, or Y91F of DHHC5-Flag/pCDH-puro and/or 0.5 μg LYN/pCDNA3.3. On day 3, cells harvested & immunoprecipitated with anti-Flag M2 beads to detect phosphorylation of DHHC5 (pY). Control & LYN knockdown adipocytes pretreated & treated with BSA or oleate (100 µM) for 1 h, followed by immunostaining (d), surface biotinylation (e), & Acyl-RAC (f) assays. Control & CD36 knockdown 3T3-L1 adipocytes pretreated & treated with oleate (100 µM) for indicated time (g) or 5 min (h). Cells harvested & subjected to immunoprecipitation of LYN to detect phosphorylation of LYN (Y396). i, j WT & Cd36-/- SVFs set up & subjected to FA uptake as in Fig. 3h, i, except that cells pretreated with PP2 (15 µM). Each value represents mean ± SEM obtained from 20 cells. Two-sided Student's t test was performed between DMSO & PP2 treated cells. k WT & Cd36-/- adipocytes set up & subjected to 3H-oleate uptake as in Fig. 3j, except that cells pretreated with PP2 (15 µM). Each value represents mean ± SEM obtained from 3samples. Two-sided Student's t test was performed between DMSO & PP2 treated cells. These experiments repeated at least twice. Source data provided as a Source Data file. Image collected & cropped by CiteAb from the following publication (https://pubmed.ncbi.nlm.nih.gov/32958780), licensed under a CC-BY license. Not internally tested by Novus Biologicals.



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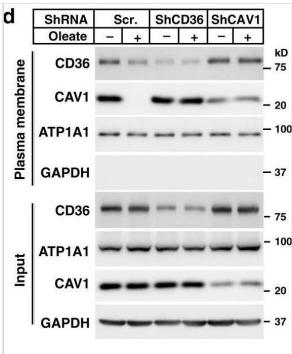


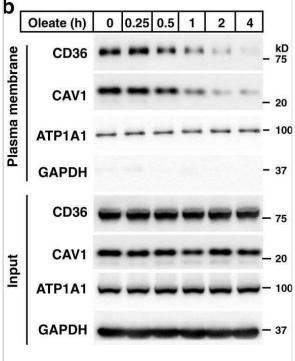
Western Blot: CD36 Antibody (1283D) - BSA Free [NBP2-54790] - FAs trigger internalization of CD36.a, b On day 8 of differentiation, 3T3-L1 adipocytes were pretreated with serum-free medium for 4 h, & then treated with BSA-conjugated oleate (100 µM) for indicated time, a One set of cells was subjected to immunostaining with anti-CD36 & anti-CAV1 antibodies. LipidTOX was used to label lipid droplets. Images were taken under a Zeiss LSM-780 microscope in a 3D Z-stack mode & reconstructed using Imaris 9.2.0. b The other set of cells was subjected to surface biotinylation assay & blotted with indicated antibodies. c, d On day 4 of differentiation, 3T3-L1 cells were infected with lentivirus encoding scrambled shRNA or shRNAs against CD36 or CAV1. On day 5, cells were selected with 5 µg/ml puromycin. On day 8, cells were pretreated as in (a) & treated with oleate (100 µM) for 4 h, followed by immunostaining with anti-CD36 & anti-CAV1 antibodies (c), or surface biotinylation assay (d). e, f 3T3-L1 adipocytes were pretreated as in (a) & treated with BSA-conjugated FAs with different chain lengths or saturation (100 µM) for 4 h. Cells were subjected to immunostaining with anti-CD36 antibody (e), or surface biotinylation assay (f). After oleate treatment for 4 h, 3T3-L1 adipocytes were switched to serum-free medium for indicated time & harvested for immunostaining (g) & surface biotinylation (h). The scale bars were as indicated in each figure. These experiments were repeated at least three times. Source data are provided as a Source Data file. Image collected & cropped by CiteAb from the following publication

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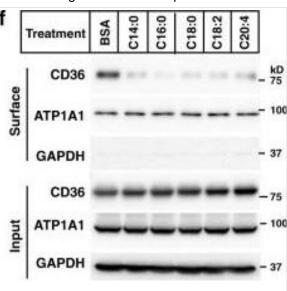
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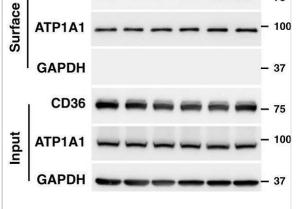


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Removal of OA (h)

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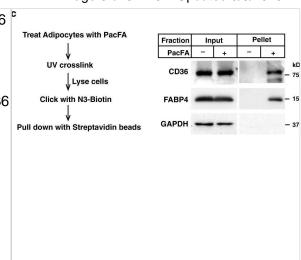
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Treatment

CD36

ATP1A1

Western Blot: CD36 Antibody (1283D) - BSA Free [NBP2-54790] - CD36 F -mediated caveolar endocytosis transports FAs into cells.a-c 3T3-L1 adipocytes were pretreated with serum-free medium for 4 h, & then treated with BSA-conjugated PacFA (50 µM) for 20 min, followed by UV crosslinking on ice for 30 min. a, b Cells were subjected to click chemistry using an N3-Alexa Fluro 488, & immunostained with anti-CD36 antibody. Colocalization of PacFA & CD36 was quantified from 24 cells over three independent experiments & plotted in (b). The value represents mean ± SEM. c Cells were lysed & subjected to click chemistry assay using N3-biotin. PacFA-labeled proteins were captured with streptavidin beads & subjected to western blot using anti-CD36 & anti-FABP4 antibodies. d WT, Cav1-/-, Cd36-/-, & Cav1-/-;Cd36-/-SVFs were isolated & differentiated into adipocytes & treated with 100 µM 3H-oleate (specific activity, 2268 dpm/nmol) for 1 h. Lipid fractions were extracted from the cells & subjected to scintillation counting. The radioactive counting was normalized to protein content. Each value represents mean ± SEM obtained from three samples. Two-sided Student's t test was performed between WT & each of the knockout cells. These experiments were repeated twice. Source data are provided as a Source Data file. Image collected & cropped by CiteAb from the following publication (https://pubmed.ncbi.nlm.nih.gov/32958780), licensed under a CC-BY license. Not internally tested by Novus Biologicals.



Publications

Hao JW, Wang J, Guo H et al. CD36 facilitates fatty acid uptake by dynamic palmitoylation-regulated endocytosis Nat Commun 2020-09-21 [PMID: 32958780] (IP)

Procedures

Western Blot Protocol for CD36/SR-B3 Antibody (NBP2-54790)

Western Blot Protocol

- 1. Perform SDS-PAGE on samples to be analyzed, loading 25 ug of total protein per lane.
- 2. Transfer proteins to membrane according to the instructions provided by the manufacturer of the membrane and transfer apparatus.
- 3. Stain according to standard Ponceau S procedure (or similar product) to assess transfer success, and mark molecular weight standards where appropriate.
- 4. Rinse the blot.
- 5. Block the membrane using standard blocking buffer for at least 1 hour.
- 6. Wash the membrane in wash buffer three times for 10 minutes each.
- 7. Dilute anti-CD36 primary antibody in blocking buffer and incubate 1 hour at room temperature.
- 8. Wash the membrane in wash buffer three times for 10 minutes each.
- 9. Apply the diluted HRP conjugated secondary antibody in blocking buffer (as per manufacturers instructions) and incubate 1 hour at room temperature.
- 10. Wash the blot in wash buffer three times for 10 minutes each (this step can be repeated as required to reduce background).
- 11. Apply the detection reagent of choice in accordance with the manufacturers instructions.

Note: Tween-20 can be added to the blocking or antibody dilution buffer at a final concentration of 0.05-0.2%.

Immunocytochemistry Protocol for CD36/SR-B3 Antibody (NBP2-54790)

Immunocytochemistry Protocol

Culture cells to appropriate density in 35 mm culture dishes or 6-well plates.

- 1. Remove culture medium and add 10% formalin to the dish. Fix at room temperature for 30 minutes.
- 2. Remove the formalin and add ice cold methanol. Incubate for 5-10 minutes.
- 3. Remove methanol and add washing solution (i.e. PBS). Be sure to not let the specimen dry out. Wash three times for 10 minutes.
- 4. To block nonspecific antibody binding incubate in 10% normal goat serum from 1 hour to overnight at room temperature.
- 5. Add primary antibody at appropriate dilution and incubate at room temperature from 2 hours to overnight at room temperature.
- 6. Remove primary antibody and replace with washing solution. Wash three times for 10 minutes.
- 7. Add secondary antibody at appropriate dilution. Incubate for 1 hour at room temperature.
- 8. Remove antibody and replace with wash solution, then wash for 10 minutes. Add Hoechst 33258 to wash solution at 1:25,0000 and incubate for 10 minutes. Wash a third time for 10 minutes.
- 9. Cells can be viewed directly after washing. The plates can also be stored in PBS containing Azide covered in Parafilm (TM). Cells can also be cover-slipped using Fluoromount, with appropriate sealing.

*The above information is only intended as a guide. The researcher should determine what protocol best meets their needs. Please follow safe laboratory procedures.





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Products Related to NBP2-54790

HAF008 Goat anti-Rabbit IgG Secondary Antibody [HRP]

NB7160 Goat anti-Rabbit IgG (H+L) Secondary Antibody [HRP]

NBP2-24891 Rabbit IgG Isotype Control

NB400-145PEP CD36 Antibody Blocking Peptide

Limitations

This product is for research use only and is not approved for use in humans or in clinical diagnosis. Primary Antibodies are guaranteed for 1 year from date of receipt.

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