

# Product Datasheet

## SREBP1 Antibody - BSA Free NB100-2215

Unit Size: 0.1 ml

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

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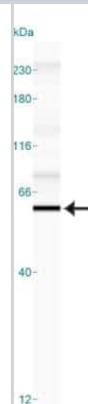
**NB100-2215**

SREBP1 Antibody - BSA Free

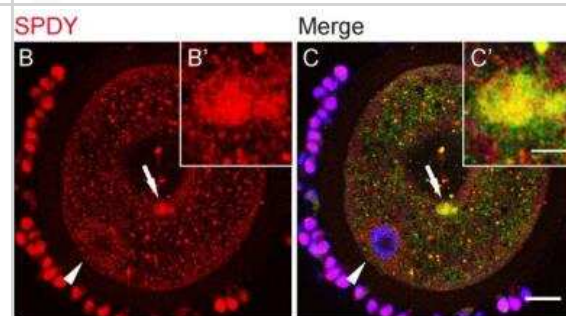
Product Information	
Unit Size	0.1 ml
Concentration	1.0 mg/ml
Storage	Store at 4C short term. Aliquot and store at -20C long term. Avoid freeze-thaw cycles.
Clonality	Polyclonal
Preservative	0.02% Sodium Azide
Isotype	IgG
Purity	Immunogen affinity purified
Buffer	PBS
Target Molecular Weight	65 kDa
Product Description	
Description	Novus Biologicals Rabbit SREBP1 Antibody - BSA Free (NB100-2215) is a polyclonal antibody validated for use in IHC, WB, ICC/IF and Simple Western. Anti-SREBP1 Antibody: Cited in 57 publications. All Novus Biologicals antibodies are covered by our 100% guarantee.
Host	Rabbit
Gene ID	6720
Gene Symbol	SREBF1
Species	Human, Mouse, Rat, Porcine, Bovine, Hamster, Plant
Reactivity Notes	Use in Human reported in scientific literature (PMID:33842305).
Immunogen	A synthetic peptide made to a portion of the human SREBP1 protein sequence (between residues 300-400). [Uniprot: P36956]
Product Application Details	
Applications	Western Blot, Simple Western, Immunohistochemistry-Paraffin, Immunocytochemistry/ Immunofluorescence, Immunohistochemistry, Microarray
Recommended Dilutions	Western Blot 1:500, Simple Western 1:25, Immunohistochemistry 1:200 - 1:500, Immunocytochemistry/ Immunofluorescence 1:200, Immunohistochemistry-Paraffin 1:200 - 1:500, Microarray reported in scientific literature (PMID 33278777)
Application Notes	<p>Unprocessed SREBP1 is an ~122 kDa integral membrane protein that moves from ER to golgi for processing. The first cleavage is performed by S2P at residue 490, and the resulting active portion is then translocated out of the golgi. This is the ~65 kDa mature form that is detected by NB100-2215. The second cleavage is performed by S1P at residue 530, and the resulting C-term portion (from residues 530-1147) remains in the golgi where it is detected as a ~65 kDa protein by NB100-60545. In Western blot, this SREBP1 antibody detects the processed form of sterol regulatory element-binding protein 1 (aa 1-490) which runs at approx. 65kDa position. In Simple Western only 10 - 15 uL of the recommended dilution is used per data point.</p> <p>See <a href="#">Simple Western Antibody Database</a> for Simple Western validation: Tested in HeLa lysate 1.0 mg/mL, separated by Size, antibody dilution of 1:25, apparent MW was 62 kDa. Separated by Size-Wes, Sally Sue/Peggy Sue.</p>

## Images

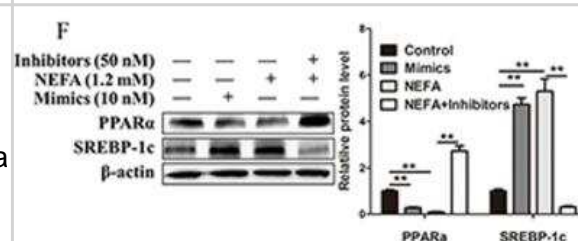
Simple Western: SREBP1 Antibody [NB100-2215] - Image shows a specific band for SREBP1 in 1 mg/mL of HeLa cell lysate. This experiment was performed under reducing conditions using the 12-230 kDa separation system.



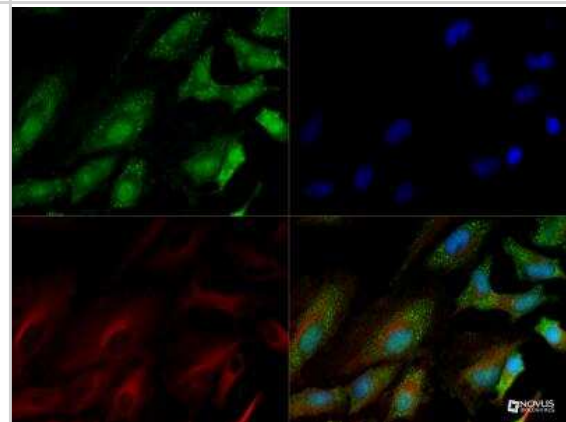
Immunocytochemistry/Immunofluorescence: SREBP1 Antibody [NB100-2215] - CDC2 associates with SPDY at ERES. (A-C) 0 h GV stage oocyte labeled for CDC2 (A, green), DNA (A, blue), and SPDY (B, red). Both CDC2 and SPDY localize to the same cortical domain (C). Note that the center of this oocyte is dented (the area that contains the arrow) causing the structure to appear in the middle of the oocyte, whereas it is located in the cortex. Images are Z-projections of 2 (A-C); scale bars represent 20  $\mu$ m in C and 5  $\mu$ m in C'. Arrows indicate the region of the oocyte that is shown enlarged in the insets (B'-C'). Arrowheads denote the position of the GV. Image collected and cropped by CiteAb from the following publication (<https://bmcdevbiol.biomedcentral.com/articles/10.1186/1471-213X-9-8>) licensed under a CC-BY license.



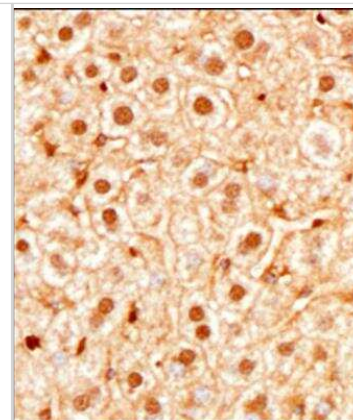
Western Blot: SREBP1 Antibody [NB100-2215] - MiR-181a overexpression impairs and miR-181a inhibition improves glucose and lipid homeostasis in HepG2 cells. HepG2 cells were divided into 4 groups as follows: a control group, mimics group (HepG2 cells transfected with 10 nM mimics), NEFA group (treated with 1.2 mM NEFA), and miR-181a + NEFA group (transfected with 50 nM miR-181a inhibitors and then treated with 1.2 mM NEFA). It was followed with or without 100 nM insulin. Immunoblot analysis (left) and quantification (right) of SREBP-1c and PPAR $\alpha$  in HepG2 cells. Image collected and cropped by Citeab from the following publication (Upregulation of miR-181a impairs hepatic glucose and lipid homeostasis. *Oncotarget* (2017)) licensed under a CC-BY license.



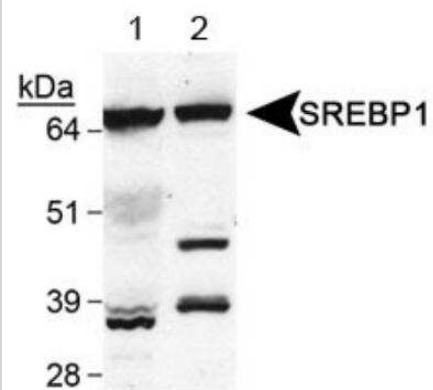
Immunocytochemistry/Immunofluorescence: SREBP1 Antibody [NB100-2215] - SREBP1 antibody was tested in HeLa cells with DyLight488 (green). Nuclei and alpha-tubulin were counterstained with DAPI (blue) and DyLight 550 (red). Nuclear and punctate vesicle staining was observed.



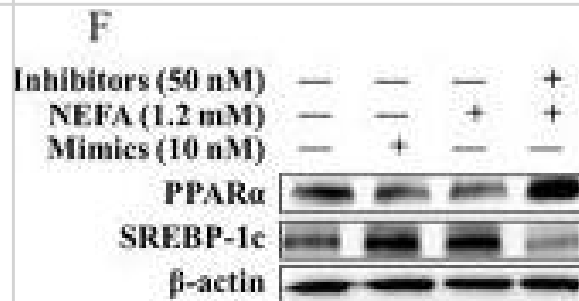
Immunohistochemistry-Paraffin: SREBP1 Antibody [NB100-2215] - analysis of FFPE tissue section of mouse liver with SREBP1 antibody at 1:200. The antibody generated an expected cytoplasmic-nuclear immunostaining of SREBP1 protein in the hepatocytes.



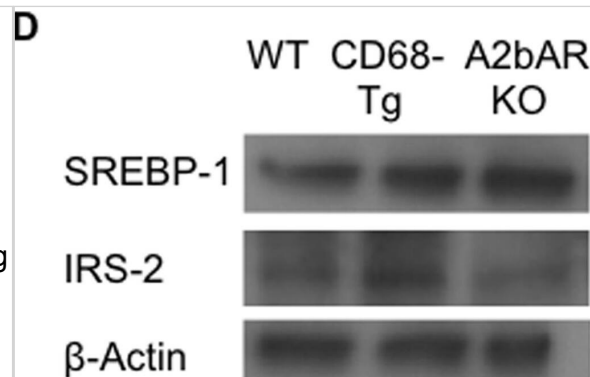
Western Blot: SREBP1 Antibody [NB100-2215] - Detection of SREBP1. Lane 1: human liver. Lane 2: mouse liver.



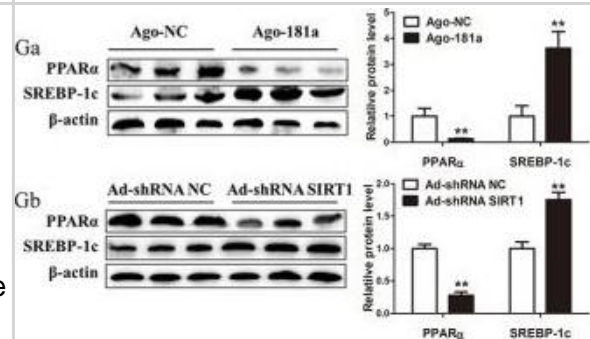
Western Blot: SREBP1 Antibody - BSA Free [NB100-2215] - MiR-181a overexpression impairs & miR-181a inhibition improves glucose & lipid homeostasis in HepG2 cells. HepG2 cells were divided into 4 groups as follows: a control group, mimics group (HepG2 cells transfected with 10 nM mimics), NEFA group (treated with 1.2 mM NEFA), & miR-181a + NEFA group (transfected with 50 nM miR-181a inhibitors & then treated with 1.2 mM NEFA). B was followed with or without 100 nM insulin. (A) Immunoblot analysis (top) & quantification (bottom) of SIRT1, PGC-1 $\alpha$  & acetylated PGC-1 $\alpha$  in HepG2 cells. (B) Immunoblotting analysis (top) & quantification (bottom) of insulin-stimulated phosphorylation of AKT in HepG2 cells. (C) Glucose concentration in medium. (D) The glycogen content in HepG2 cells. (E) The mRNA expression levels of G6Pase & PEPCK. (F) Immunoblot analysis (left) & quantification (right) of SREBP-1c & PPAR $\alpha$  in HepG2 cells. (G) TG content in HepG2 cells. \*P < 0.05, \*\*P < 0.01. All experiments were repeated at least three times & representative results are shown. Image collected & cropped by CiteAb from the following publication (<https://pubmed.ncbi.nlm.nih.gov/29207650>), licensed under a CC-BY license. Not internally tested by Novus Biologicals.



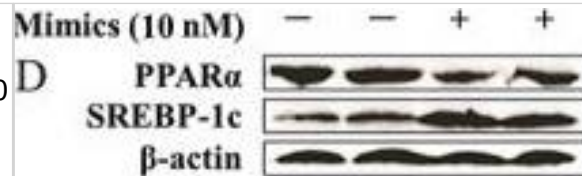
Western Blot: SREBP1 Antibody - BSA Free [NB100-2215] - Effect of restoration of macrophage A2bAR on liver. Liver was collected from WT, A2bAR KO, & CD68-Tg mice after 16 weeks of HFD as described in the methods. Relative mRNA expression was determined using the  $\Delta\Delta CT$  method with normalization to 18s rRNA. A. mRNA expression of TNF- $\alpha$  in liver. A2bAR KO (n=5) vs WT (n=7) p-value =0.0271; A2bAR KO vs CD68-Tg (n=6) p-value =0.0210. B. mRNA expression of IL-6 in liver. A2bAR KO (n=8) vs WT (n=7) p-value =0.0159; A2bAR KO vs CD68-Tg (n=8) p-value =0.0080. C. mRNA expression of IRS-2 in liver. CD68-Tg (n=7) vs A2bAR KO (n=8) p-value =0.0300. D. Western blot analysis of liver; one representative (of 3 sets) WT, CD68-Tg & A2bAR KO group shown at 15 minutes post-insulin injection, following 16 weeks of HFD. Levels of mature SREBP-1 (68 kDa), & IRS-2 (185 kDa), were probed by Western blot analysis, using  $\beta$ -actin (43 kDa) as loading control. Quantification of Western Blot results was performed with Image J software (<http://rsb.info.nih.gov/ij/>) with normalization to  $\beta$ -actin. WT to A2bAR KO: IRS-2 p-value =0.0092, SREBP-1 p-value =0.0154; CD68-Tg to A2bAR KO: IRS-2 p-value =0.0247, SREBP-1 p-value =0.0170. Data are averages  $\pm$  SD. \*Student two-tail t-test assuming equal variance was found significant only when p-value <0.05. Image collected & cropped by CiteAb from the following publication (<https://pubmed.ncbi.nlm.nih.gov/24892847>), licensed under a CC-BY license. Not internally tested by Novus Biologicals.



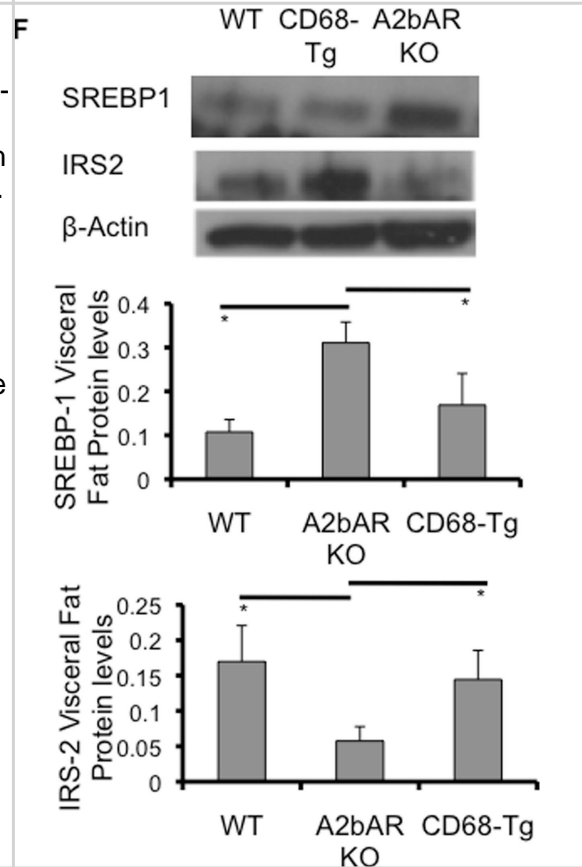
Western Blot: SREBP1 Antibody - BSA Free [NB100-2215] - MiR-181a overexpression or SIRT1 knockdown impairs glucose & lipid metabolism in vivo (Aa & Ab) Immunoblotting analysis (left) & quantification (right) of SIRT1, PGC-1 $\alpha$  & acetylated PGC-1 $\alpha$  in the liver of mice injected with Ago-181a or Ago-NC or Ad-shRNA NC or Ad-shRNA SIRT1. (B) Glucose tolerance test of mice injected with Ago-181a (n = 7) or Ago-NC (n = 7) or Ad-shRNA NC (n = 6) or Ad-shRNA SIRT1 (n = 6). (C) Insulin tolerance test of mice injected with Ago-181a (n = 7) or Ago-NC (n = 7) or Ad-shRNA NC (n = 6) or Ad-shRNA SIRT1 (n = 6). (D) Representative images of PAS staining (original magnification  $\times$  20) of liver sections from mice injected with Ago-181a or Ago-NC or Ad-shRNA NC or Ad-shRNA SIRT1. (Ea & Eb) Immunoblotting analysis (top) & quantification (bottom) of insulin-stimulated phosphorylation of IR, AKT & GSK3 $\beta$  in liver of mice injected with Ago-181a or Ago-NC or Ad-shRNA NC or Ad-shRNA SIRT1. (F) Representative images of H&E staining (left, original magnification  $\times$  20) & Oil-red staining (right, original magnification  $\times$  20) of liver sections from mice injected with Ago-181a or Ago-NC or Ad-shRNA NC or Ad-shRNA SIRT1. (Ga & Gb) Immunoblotting analysis (top) & quantification (bottom) of SREBP-1c & PPAR $\alpha$  in the liver of mice injected with Ago-181a or Ago-NC or Ad-shRNA NC or Ad-shRNA SIRT1. \*P < 0.05, \*\*P < 0.01. All experiments were repeated at least three times & representative results are shown. Image collected & cropped by CiteAb from the following publication (<https://pubmed.ncbi.nlm.nih.gov/29207650>), licensed under a CC-BY license. Not internally tested by Novus Biologicals.



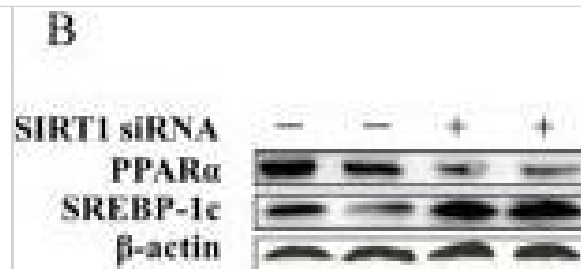
Western Blot: SREBP1 Antibody - BSA Free [NB100-2215] - MiR-181a overexpression impairs & miR-181a inhibition improves glucose & lipid metabolism in vitro(A, B, C, D & E) Hepatocytes were transfected with 10 nM miR-181a mimics or negative controls in the absence or presence of 100 nM insulin. (A) Immunoblot analysis (left) & quantification (right) of insulin-stimulated phosphorylation of IR, Akt & GSK3 $\beta$  in hepatocytes. (B) The mRNA expression levels of G6Pase & PEPCK in hepatocytes. (C) Glucose concentration in medium. (D) Immunoblot analysis (left) & quantification (right) of SREBP-1c & PPAR $\alpha$  expression in hepatocytes. (E) TG content in hepatocytes. (F, G, H, I & J) Hepatocytes were transfected with 50 nM miR-181a inhibitors or negative controls in the absence or presence of 100 nM insulin. (F) Immunoblot analysis (left) & quantification (right) of insulin-stimulated phosphorylation of IR, Akt & GSK3 $\beta$  in hepatocytes. (G) The expression levels of G6Pase & PEPCK in hepatocytes. (H) Glucose concentration in medium. (I) Immunoblot analysis (left) & quantification (right) of SREBP-1c & PPAR $\alpha$  expression in hepatocytes. (J) TG content in hepatocytes. \*P < 0.05, \*\*P < 0.01. All experiments were repeated at least three times & representative results are shown. Image collected & cropped by CiteAb from the following publication (<https://pubmed.ncbi.nlm.nih.gov/29207650>), licensed under a CC-BY license. Not internally tested by Novus Biologicals.



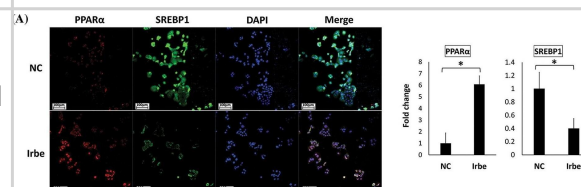
Western Blot: SREBP1 Antibody - BSA Free [NB100-2215] - Effect of restoration of macrophage A2bAR on adipose tissue. Visceral (epididymal) adipose tissue was collected from WT, A2bAR KO, & CD68-Tg mice after 16 weeks of HFD as described in the methods. A-C,E. Relative mRNA expression was determined using the  $\Delta\Delta\text{CT}$  method with normalization to 18s rRNA. A. mRNA expression of TNF- $\alpha$  in visceral fat. A2bAR KO (n=7) vs CD68-Tg (n=9) p-value = 0.0345. B. mRNA expression of IL-6 in visceral fat. A2bAR KO (n=7) vs WT (n=7) p-value = 0.0073; A2bAR KO vs CD68-Tg (n=7) p-value = 0.0141. C. mRNA expression of MCP1 in visceral fat. A2bAR KO (n=7) vs CD68-Tg (n=9) p-value = 0.0232. D. Visceral adipose tissue from WT, A2bAR KO, & CD68-Tg mice after 16 weeks of HFD was fixed in 4% paraformaldehyde & paraffin-embedded. Sections immunostained with the macrophage marker F4/80. Representative sections for each genotype at a magnification of 200x & 400x. Arrows point to crown-like structures. E. mRNA expression of IRS-2 in visceral fat. CD68-Tg (n=8) vs A2bAR KO (n=8) p-value = 0.0115, WT (n=6) vs A2bAR KO p-value = 0.0358. F. Western blot analysis of visceral fat; one representative (of 3 sets) WT, CD68-Tg & A2bAR KO group shown at 15 minutes post-insulin injection, following 16 weeks of HFD. WT to A2bAR KO: IRS-2 p-value = 0.0411, SREBP-1 p-value = 0.0103. CD68-Tg to A2bAR KO: IRS-2 p-value = 0.0305, SREBP-1 p-value = 0.0459. Data are averages  $\pm$  SD. \*Student two-tail t-test assuming equal variance was found significant only when p-value < 0.05. Image collected & cropped by CiteAb from the following publication (<https://pubmed.ncbi.nlm.nih.gov/24892847>), licensed under a CC-BY license. Not internally tested by Novus Biologicals.



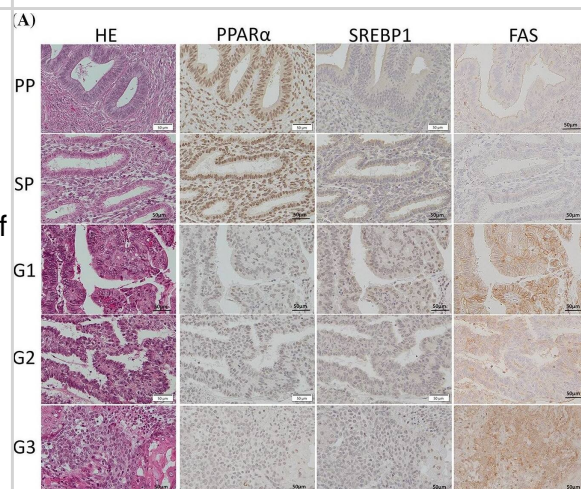
Western Blot: SREBP1 Antibody - BSA Free [NB100-2215] - SIRT1 knockdown impairs & SIRT1 overexpression improves glucose & lipid metabolism in hepatocytes(A) Immunoblot analysis (left) & quantification (right) of insulin-stimulated phosphorylation of IR, Akt & GSK3 $\beta$  protein levels in hepatocytes. Hepatocytes were transfected with SIRT1 siRNA with or without 100 nM insulin. (B) Immunoblot analysis (left) & quantification (right) of SREBP-1c & PPAR $\alpha$  protein levels in hepatocytes. Hepatocytes were transfected with SIRT1 siRNA. (C) Immunoblot analysis (left) & quantification (right) of SIRT1, PGC-1 $\alpha$  & acetylated PGC-1 $\alpha$  in hepatocytes. Hepatocytes were infected with Ad-SIRT1. (D) Immunoblotting analysis (left) & quantification (right) of insulin-stimulated phosphorylation of IR, AKT & GSK3 $\beta$  in hepatocytes. Hepatocytes were treated with 1.2 mM NEFA or 1.2 mM NEFA & Ad-SIRT1 with or without 100 nM insulin. (E, F & G) Hepatocytes were treated with 1.2 mM NEFA or 1.2 mM NEFA & Ad-SIRT1. (E) Glucose concentration in medium. (F) Immunoblot analysis (top) & quantification (bottom) of SREBP-1c & PPAR $\alpha$  in hepatocytes. (G) TG contents in hepatocytes. \*P < 0.05, \*\*P < 0.01. All experiments were repeated at least three times & representative results are shown. Image collected & cropped by CiteAb from the following publication (<https://pubmed.ncbi.nlm.nih.gov/29207650>), licensed under a CC-BY license. Not internally tested by Novus Biologicals.



The PPAR $\alpha$  activator, Irbe, reduces SREBP1 expression and induces the expression and co-localization of PPAR $\alpha$  and p21/p27 in Ishikawa cells. (A) Dual immunofluorescent staining for PPAR $\alpha$  (red) and SREBP1 (green) show that the Irbe (100  $\mu$ M) treatment increased the nuclear expression of the PPAR $\alpha$  protein while decreasing that of SREBP1. (B, C) Dual immunofluorescent staining for PPAR $\alpha$  (red) and p21 (B) or p27 (C) (green) showed that the Irbe treatment increased the expression of both proteins, and their distribution was almost consistent. \*p < 0.05. Image collected and cropped by CiteAb from the following open publication (<https://pubmed.ncbi.nlm.nih.gov/37305395>), licensed under a CC-BY license. Not internally tested by Novus Biologicals.



PPAR $\alpha$  expression was lower and SREBP1 and FAS expression was higher in endometrial carcinoma (EMC) than in normal endometrial glands. (Immunohistochemistry). (A) Photomicrographs show hematoxylin and eosin (HE) staining and representative immunostaining for PPAR $\alpha$ , SREBP1, and FAS in serial sections of the normal endometrium (PP: proliferation phase; SP: secretory phase) and EMC (G1: grade 1; G2: grade 2; G3: grade 3). (B–D) Graphic demonstration of immunostaining for PPAR $\alpha$  (B), SREBP1 (C), and FAS (D). The expression of PPAR $\alpha$  was significantly lower in EMC than in the normal endometrium, while the expression of SREBP1 and FAS was significantly higher (\* p < 0.01). (E) The graph indicates the H-scores of PPAR $\alpha$ , SREBP1, and FAS in non-obese EMC patients with BMI <25 (n = 26) or in obese EMC patients with BMI >25 (n = 24). There was no significant difference. (F) The BMI and H-scores of PPAR $\alpha$ , SREBP1, and FAS in each EMC patient are shown in scatter plots. Spearman's rank correlation coefficient showed a weak inverse correlation between BMI and the PPAR $\alpha$  H-score and a moderate or weak correlation between BMI and SREBP1 or FAS. Image collected and cropped by CiteAb from the following open publication (<https://pubmed.ncbi.nlm.nih.gov/37305395>), licensed under a CC-BY license. Not internally tested by Novus Biologicals.



## Publications

Li J, Zhao C, Zu K et al. Salidroside protects bovine hepatocytes against fatty acid-induced lipid accumulation and inflammation by activating AMPK/SIRT1 pathway. *Journal of dairy science* 2025-07-09 [PMID: 40645487]

Senatus L, Egaña-Gorroño L, López-Díez R et al. DIAPH1 mediates progression of atherosclerosis and regulates hepatic lipid metabolism in mice *Communications Biology* 2023-03-17 [PMID: 36932214]

Zhang B, Yang W, Wang S et al. Lipid Accumulation and Injury in Primary Calf Hepatocytes Challenged With Different Long-Chain Fatty Acids *Frontiers in Veterinary Science* 2020-10-15 [PMID: 33195520]

Xue C, Zeng P, Gong K et al. Nogo-B inhibition facilitates cholesterol metabolism to reduce hypercholesterolemia *Cell Rep* 2024-09-03 [PMID: 39235944]

Li M, Zhao B, Wang J et al. Caveolin 1 in bovine liver is associated with fatty acid-induced lipid accumulation and the ER unfolded protein response: role in fatty liver development. *Journal of dairy science* 2024-09-27 [PMID: 39343220]

Ladraa S, Zerbib L, Bayard C et al. PIK3CA gain-of-function mutation in adipose tissue induces metabolic reprogramming with Warburg-like effect and severe endocrine disruption *Science advances* 2022-12-09 [PMID: 36490341]

Zhu Y, Lei L, Wang X et al. Low abundance of insulin-induced gene 1 contributes to SREBP-1c processing and hepatic steatosis in dairy cows with severe fatty liver *Journal of dairy science* 2023-06-06 [PMID: 37291038]

John G. Purdy, Thomas Shen, Joshua D. Rabinowitz Fatty Acid Elongase 7 Catalyzes the Lipidome Remodeling Essential for Human Cytomegalovirus Replication *Cell reports* 2015-02-25 [PMID: 25732827]

TS Huang, T Wu, YD Wu, XH Li, J Tan, CH Shen, SJ Xiong, ZQ Feng, SF Gao, H Li, WB Cai Long-term statins administration exacerbates diabetic nephropathy via ectopic fat deposition in diabetic mice *Nature Communications*, 2023-01-24;14(1):390. 2023-01-24 [PMID: 36693830]

Melissa L. Erickson, Zachary W. Patinkin, Allison M. Duensing, Dana Dabelea, Leanne M. Redman, Kristen E. Boyle Maternal metabolic health drives mesenchymal stem cell metabolism and infant fat mass at birth *JCI Insight* 2021-07-08 [PMID: 34061777]

Aleksandra Aizenshtadt, Chencheng Wang, Shadab Abadpour, Pedro Duarte Menezes, Ingrid Wilhelmsen, Andrea Dalmao-Fernandez, Justyna Stokowiec, Alexey Golovin, Mads Johnsen, Thomas M D Combriat, Hanne Røberg-Larsen, Nikolaj Gadegaard, Hanne Scholz, Mathias Busek, Stephan J K Krauss Pump-Less, Recirculating Organ-On-Chip (rOoC) Platform To Model The Metabolic Crosstalk Between Islets and Liver. *Advanced healthcare materials* 2024-01-14 [PMID: 38221504]

Shiota T, Li Z, Chen GY et al. Hepatoviruses promote very-long-chain fatty acid and sphingolipid synthesis for viral RNA replication and quasi-enveloped virus release *Science advances* 2023-10-20 [PMID: 37862421] (WB, Human)

Details:

1:1000 WB dilution

More publications at <http://www.novusbio.com/NB100-2215>

## Procedures

### Western Blot protocol for SREBP1 Antibody (NB100-2215)

#### Western Blot Protocol

1. Perform SDS-PAGE (4-12%) on samples to be analyzed, loading 35 ug of total protein per lane.
2. Transfer proteins to Nitrocellulose according to the instructions provided by the manufacturer of the transfer apparatus.
3. Rinse membrane with dH<sub>2</sub>O and then stain the blot using ponceau S for 1-2 minutes to access the transfer of proteins onto the nitrocellulose membrane. Rinse the blot in water to remove excess stain and mark the lane locations and locations of molecular weight markers using a pencil.
4. Rinse the blot in TBS for approximately 5 minutes.
5. Block the membrane using 5% non-fat dry milk + 1% BSA in TBS for 1 hour at room temperature (RT).
6. Rinse the membrane in dH<sub>2</sub>O and then wash the membrane in wash buffer [TBS + 0.1% Tween] 3 times for 10 minutes each.
7. Dilute the rabbit anti-SREBP1 primary antibody (NB 100-2215) in blocking buffer and incubate 1 hour at RT.
8. Rinse the membrane in dH<sub>2</sub>O and then wash the membrane in wash buffer [TBS + 0.1% Tween] 3 times for 10 minutes each.
9. Apply the diluted rabbit-IgG HRP-conjugated secondary antibody in blocking buffer (as per manufacturer's instructions) and incubate 1 hour at room temperature.
10. Wash the blot in wash buffer [TBS + 0.1% Tween] 3 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 manufacturer's instructions (we used BioFX Super Plus ECL).

Note: Tween-20 can be added to the blocking or antibody dilution buffer at a final concentration of 0.05-0.2%, provided it does not interfere with antibody-antigen binding.



**Immunocytochemistry/Immunofluorescence protocol for SREBP1 Antibody (NB100-2215)**

## 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 1:25,000 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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Fax: 905.827.6402  
canada.inquires@bio-techne.com

### **Bio-Techne Ltd**

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### **Products Related to NB100-2215**

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NB820-59662	Mouse Liver Whole Tissue Lysate (Adult Whole Normal)
NB100-2215PEP	SREBP1 Antibody Blocking Peptide
NBP2-33376H	Blue Marker Antibody (6F4-F6) [HRP]
HAF008	Goat anti-Rabbit IgG Secondary Antibody [HRP]
NB7160	Goat anti-Rabbit IgG (H+L) Secondary Antibody [HRP]
NBP2-24891	Rabbit IgG Isotype Control

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### **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.

For more information on our 100% guarantee, please visit [www.novusbio.com/guarantee](http://www.novusbio.com/guarantee)

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