Product Datasheet

SUCNR1/GPR91 Antibody - BSA Free NBP1-00861

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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NBP1-00861

SUCNR1/GPR91 Antibody - BSA Free

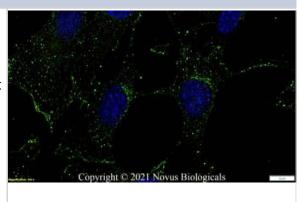
Product Information	
Unit Size	0.1 mg
Concentration	1.0 mg/ml
Storage	Store at 4C short term. Aliquot and store at -20C long term. Avoid freeze-thaw cycles.
Clonality	Polyclonal
Preservative	0.02% Sodium Azide
Isotype	IgG
Purity	Immunogen affinity purified
Buffer	PBS
Target Molecular Weight	38 kDa
Product Description	
Description	Novus Biologicals Rabbit SUCNR1/GPR91 Antibody - BSA Free (NBP1-00861) is a polyclonal antibody validated for use in IHC, WB and ICC/IF. Anti-

Product Description	
Description	Novus Biologicals Rabbit SUCNR1/GPR91 Antibody - BSA Free (NBP1-00861) is a polyclonal antibody validated for use in IHC, WB and ICC/IF. Anti-SUCNR1/GPR91 Antibody: Cited in 23 publications. All Novus Biologicals antibodies are covered by our 100% guarantee.
Host	Rabbit
Gene ID	56670
Gene Symbol	SUCNR1
Species	Human, Mouse, Rat
Specificity/Sensitivity	GPR91 (A135) detects endogenous levels of GPR91 protein.
Immunogen	A synthetic peptide made to an internal portion of the human GPR91 protein (between residues 100-200) [Uniprot: Q9BXA5]

	(
Product Application Details	
Applications	Western Blot, Immunohistochemistry-Paraffin, Immunocytochemistry/ Immunofluorescence, Immunohistochemistry, In vitro assay
Recommended Dilutions	Western Blot 1-2 ug/ml, Immunohistochemistry 1:200, Immunocytochemistry/ Immunofluorescence 1:50-1:200, Immunohistochemistry-Paraffin 1:200, In vitro assay reported in scientific literature (PMID 30478422)

Images

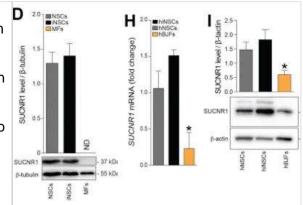
Immunocytochemistry/Immunofluorescence: SUCNR1/GPR91 Antibody [NBP1-00861] - NIH3T3 cells were fixed in 4% paraformaldehyde for 10 minutes and permeabilized in 0.05% Triton X-100 in PBS for 5 minutes. The cells were incubated with anti-SUCNR1/GPR91 Antibody NBP1-00861 at 2 ug/ml overnight at 4C and detected with an anti-rabbit Dylight 488 (Green) at a 1:1000 dilution for 60 minutes. Nuclei were counterstained with DAPI (Blue). Cells were imaged using a 100X objective and digitally deconvolved.





Western Blot: SUCNR1/GPR91 Antibody [NBP1-00861] - D) SUCNR1 protein expression relative to B-tubulin in vitro. Data are shown as mean (+/- SEM) of n >= 3 independent replicates per condition. (H) qRT-PCR of SUCNR1 basal expression in human cells. Data are normalized on 18S and expressed as mean fold change (+/- SEM) versus NSCs from n >= 3 independent replicates per condition. (I) Representative blot of SUNCR1 basal protein expression in human cells. hBJFs, human BJ fibroblasts. *p <= 0.05 versus 0'. Image collected and cropped by CiteAb from the following publication

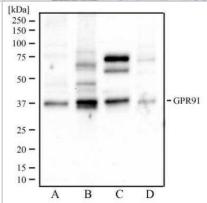
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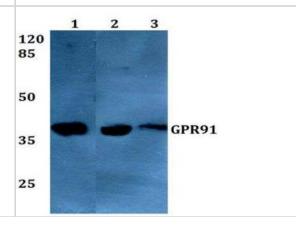
Immunohistochemistry-Paraffin: SUCNR1/GPR91 Antibody [NBP1-00861] - Staining of GPR91 in mouse Vas Deferens where strong membrane staining is observed.



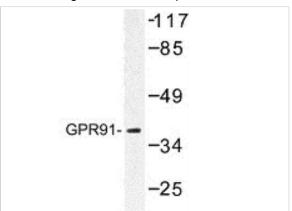
Western Blot: SUCNR1/GPR91 Antibody [NBP1-00861] - Analysis of human kidney tissue (A), mouse kidney tissue (B), rat kidney tissue (C), and hek293 cells (D) using GPR91 antibody at 2ug/ml.



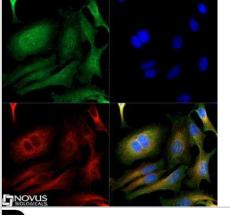
Western Blot: SUCNR1/GPR91 Antibody [NBP1-00861] - Lane1:Hela whole cell lysate. Lane2:Mouse kidney tissue lysate. Lane3:Rat kidney tissue lysate.



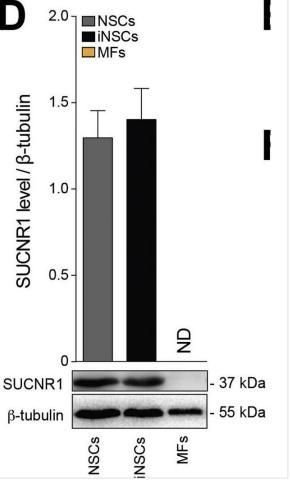
Western Blot: SUCNR1/GPR91 Antibody [NBP1-00861] - Analysis of GPR91 (A135) antibody in extracts from HUVEC/MCF-7 cells.



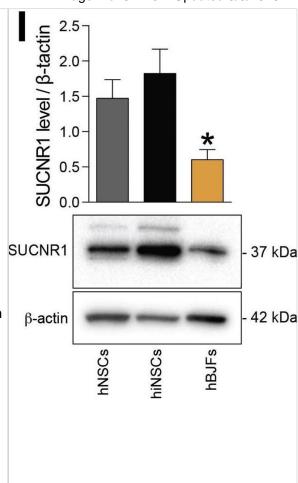
Immunocytochemistry/Immunofluorescence: SUCNR1/GPR91 Antibody [NBP1-00861] - GPR91 antibody (1:50) was tested in HeLa cells with Dylight 488 (green). Nuclei and alpha-tubulin were counterstained with DAPI (blue) and Dylight 550 (red). Image objective 40x.



Western Blot: SUCNR1/GPR91 Antibody - BSA Free [NBP1-00861] -Succinate Signals via SUCNR1 in Mouse & Human NSCs(A-C) Representative confocal microscopy images of meningeal perivascular areas with transplanted fGFP+ iNSCs (A) & NSCs (B) expressing SUCNR1 in the brain of a mouse with EAE. The image in (C) shows transplanted SUCNR1+ iNSCs in close vicinity to SUCNR1+/F4/80+ MPs. Nuclei are stained with DAPI.(D) SUCNR1 protein expression relative to β -tubulin in vitro. Data are shown as mean (\pm SEM) of $n \ge 3$ independent replicates per condition.(E) Experimental setup for succinate treatment of iNSCs/NSCs in vitro.(F) Intracellular Ca2+ response after treatment with 500 µM succinate (live staining with Fluo-4AM). Representative images (baseline & during stimulation) are pseudocolored with red/blue according to high/low fluorescence intensity. Data are mean changes in fluorescence intensity as ΔF/F0 (±SEM) from n ≥ 3 experiments.(G) Phospho-p38 MAPK (P-p38) & total p38 MAPK (p38) protein expression after succinate treatment. Data are P-p38/p38 expression relative to β-tubulin & expressed as mean fold change (±SEM) versus untreated cells over n ≥ 3 independent experiments per condition.(H) gRT-PCR of SUCNR1 basal expression in human cells. Data are normalized on 18S & expressed as mean fold change (±SEM) versus NSCs from n ≥ 3 independent replicates per condition.(I) Representative blot of SUNCR1 basal protein expression in human cells.(J) P-p38 & p38 protein expression after stimulation with succinate ± pre-treatment with the irreversible inhibitor of the human SUCNR1 4c. The scale bars represent 25 μ m. $\Box p \le 0.05$ versus 0'. hBJFs, human BJ fibroblasts; ND, not detected. See also Figure S4. Image collected & cropped by CiteAb from the following publication (https://pubmed.ncbi.nlm.nih.gov/29478844), licensed under a CC-BY license. Not internally tested by Novus Biologicals.



Western Blot: SUCNR1/GPR91 Antibody - BSA Free [NBP1-00861] -Succinate Signals via SUCNR1 in Mouse & Human NSCs(A-C) Representative confocal microscopy images of meningeal perivascular areas with transplanted fGFP+ iNSCs (A) & NSCs (B) expressing SUCNR1 in the brain of a mouse with EAE. The image in (C) shows transplanted SUCNR1+ iNSCs in close vicinity to SUCNR1+/F4/80+ MPs. Nuclei are stained with DAPI.(D) SUCNR1 protein expression relative to β -tubulin in vitro. Data are shown as mean (\pm SEM) of n \geq 3 independent replicates per condition.(E) Experimental setup for succinate treatment of iNSCs/NSCs in vitro.(F) Intracellular Ca2+ response after treatment with 500 µM succinate (live staining with Fluo-4AM). Representative images (baseline & during stimulation) are pseudocolored with red/blue according to high/low fluorescence intensity. Data are mean changes in fluorescence intensity as ΔF/F0 (\pm SEM) from n \geq 3 experiments.(G) Phospho-p38 MAPK (P-p38) & total p38 MAPK (p38) protein expression after succinate treatment. Data are P-p38/p38 expression relative to β-tubulin & expressed as mean fold change (±SEM) versus untreated cells over n ≥ 3 independent experiments per condition.(H) qRT-PCR of SUCNR1 basal expression in human cells. Data are normalized on 18S & expressed as mean fold change (±SEM) versus NSCs from n ≥ 3 independent replicates per condition.(I) Representative blot of SUNCR1 basal protein expression in human cells.(J) P-p38 & p38 protein expression after stimulation with succinate ± pre-treatment with the irreversible inhibitor of the human SUCNR1 4c. The scale bars represent 25 μ m. $\Box p \le 0.05$ versus 0'. hBJFs, human BJ fibroblasts; ND, not detected. See also Figure S4. Image collected & cropped by CiteAb from the following publication (https://pubmed.ncbi.nlm.nih.gov/29478844), licensed under a CC-BY license. Not internally tested by Novus Biologicals.





Publications

Yu W, Moninger TO, Rector MV et al. Pulmonary neuroendocrine cells sense succinate to stimulate myoepithelial cell contraction Developmental Cell 2022-09-14 [PMID: 36108628] (Western Blot, Immunocytochemistry/Immunofluorescence, Rat)

Y Guo, F Xu, SC Thomas, Y Zhang, B Paul, S Sakilam, S Chae, P Li, C Almeter, AR Kamer, P Arora, DT Graves, D Saxena, X Li Targeting the succinate receptor effectively inhibits periodontitis Cell Reports, 2022-09-20;40 (12):111389. 2022-09-20 [PMID: 36130514] (Western Blot, Immunocytochemistry/ Immunofluorescence, Rat)

Tianyi Shen, Ruoyi Lin, Chengyu Hu, Donghui Yu, Chengda Ren, Tingting Li, Meijiang Zhu, Zhongqi Wan, Tu Su, Yan Wu, Wenting Cai, Jing Yu Succinate-induced macrophage polarization and RBP4 secretion promote vascular sprouting in ocular neovascularization Journal of Neuroinflammation 2023-12-21 [PMID: 38129891]

Pu M, Zhang J, Hong F et al. The pathogenic role of Succinate-SUCNR1: A critical function that induces renal fibrosis via infiltration of M2 macrophage Research Square 2023-08-21 (WB, Mouse)

Cao Z, Mu S, Wang M et al. Succinate pretreatment attenuates intestinal ischemia-reperfusion injury by inhibiting necroptosis and inflammation via upregulating Klf4 International immunopharmacology 2023-07-01 [PMID: 37285681] (WB, Mouse)

Xu J, Tian Z, Li Z et al. Puerarin-Tanshinone IIA Suppresses atherosclerosis inflammatory plaque via targeting succinate/HIF-1alpha/IL-1beta axis Journal of ethnopharmacology 2023-05-29 [PMID: 37257708] (WB, Mouse)

Wang Y, Tao H, Tang W et al. Succinate level is increased and succinate dehydrogenase exerts forward and reverse catalytic activities in lipopolysaccharides-stimulated cardiac tissue: The protective role of dimethyl malonate European journal of pharmacology 2022-12-20 [PMID: 36549501] (B/N, Mouse)

Gudgeon N, Munford H, Bishop EL et al. Succinate uptake by T cells suppresses their effector function via inhibition of mitochondrial glucose oxidation Cell reports 2022-08-16 [PMID: 35977513] (WB)

Details:

Dilutions: 1:500

Sanchez M, Hamel D, Bajon E et al. The Succinate Receptor SUCNR1 Resides at the Endoplasmic Reticulum and Relocates to the Plasma Membrane in Hypoxic Conditions Cells 2022-07-13 [PMID: 35883628] (IHC-P, Human)

Xu G, Yuan Y, Luo P et al. Acute Succinate Administration Increases Oxidative Phosphorylation and Skeletal Muscle Explosive Strength via SUCNR1 Frontiers in veterinary science 2022-01-14 [PMID: 35097053] (IF/IHC, WB, Mouse)

Moyon A, Garrigue P, Balasse L et al. Succinate Injection Rescues Vasculature and Improves Functional Recovery Following Acute Peripheral Ischemia in Rodents: A Multimodal Imaging Study Cells 2021-04-02 [PMID: 33918298] (IF/IHC, WB, ICC/IF, Mouse)

Liu P, Wang J, Wen W et al. Cinnamaldehyde suppresses NLRP3 derived IL-1 beta via activating succinate/HIF-1 in rheumatoid arthritis rats Int. Immunopharmacol. 2020-01-01 [PMID: 32413739] (IF/IHC, Rat)

More publications at http://www.novusbio.com/NBP1-00861



Procedures

Western Blot protocol for SUCNR1/GPR91 Antibody (NBP1-00861)

Western Blot Protocol

- 1. Perform SDS-PAGE with a 12% gel 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-GPR91 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%.

Immunohistochemistry-Paraffin protocol for SUCNR1/GPR91 Antibody (NBP1-00861)

Immunohistochemistry-Paraffin Embedded Sections

Antigen Unmasking:

Bring slides to a boil in 10 mM sodium citrate buffer (pH 6.0) then maintain at a sub-boiling temperature for 10 minutes. Cool slides on bench-top for 30 minutes.

Staining:

- 1. Wash sections in deionized water three times for 5 minutes each.
- 2. Wash sections in wash buffer for 5 minutes.
- 3. Block each section with 100-400 ul blocking solution for 1 hour at room temperature.
- 4. Remove blocking solution and add 100-400 ul diluted primary antibody. Incubate overnight at 4 C.
- 5. Remove antibody solution and wash sections in wash buffer three times for 5 minutes each.
- 6. Add 100-400 ul biotinylated diluted secondary antibody. Incubate 30 minutes at room temperature.
- 7. Remove secondary antibody solution and wash sections three times with wash buffer for 5 minutes each.
- 8. Add 100-400 ul Streptavidin-HRP reagent to each section and incubate for 30 minutes at room temperature.
- 9. Wash sections three times in wash buffer for 5 minutes each.
- 10. Add 100-400 ul DAB substrate to each section and monitor staining closely.
- 11. As soon as the sections develop, immerse slides in deionized water.
- 12. Counterstain sections in hematoxylin.
- 13. Wash sections in deionized water two times for 5 minutes each.
- 14. Dehydrate sections.
- 15. Mount coverslips.



Immunocytochemistry/Immunofluorescence protocol for SUCNR1/GPR91 Antibody (NBP1-00861)

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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