Product Datasheet

Ppara Antibody (orb345414)

Catalog Number	orb345414
Category	Antibodies
Description	PPAR alpha antibody
Target	Ppara
Clonality	Polyclonal
Species/Host	Rabbit
Isotype	IgG
Conjugation	Unconjugated
Reactivity	Human, Mouse
Form/Appearance	Liquid (sterile filtered)
Concentration	1.0 mg/mL
Buffer/Preservatives	Preservative: 0.01% (w/v) Sodium Azide. Stabilizer: None; Buffer: 0.02 M Potassium Phosphate, 0.15 M Sodium Chloride, pH 7.2
Purity	This affinity purified antibody is directed against mouse PPAR alpha protein. The product was affinity purified from monospecific antiserum by immunoaffinity purification. A BLAST analysis was used to suggest reactivity with this protein from mouse, rat, bovine, dog, golden hamster and boar sources based on 100% homology for the immunogen sequence. Cross reactivity with PPAR alpha protein from human, chimpanzee and rhesus monkey may also occur as this sequence shows 88% homology (16/18 identities) with the protein from these sources. Cross reactivity with PPAR alpha homologues from other sources has not been determined. No reactivity is expected against other subtypes of PPAR.

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Immunogen	PPAR alpha Antibody was prepared from whole rabbit serum produced by repeated immunizations with a synthetic peptide corresponding to a N-Terminal region near amino acids 1-25 of mouse PPAR alpha.
UniProt ID	**P23204**
Tested applications	ELISA, IF, IHC, WB
Dilution range	ELISA: 1:8,000 - 1:32,000, IHC: 1:100-1:300, WB: 1:500 - 1:2,000
Application notes	Anti-PPAR alpha Antibody has been tested in ELISA, Western Blot, Immunohistochemistry, and Immunofluorescence. Expect a single band approximately 52 kDa in size corresponding to PPAR alpha by western blot in the appropriate tissue or cell lysate. A 1:200 dilution is suggested for Immunohistochemistry. Specific conditions for reactivity should be optimized by the end user.
Antibody Type	Primary Antibody
Storage	Store vial at -20° C or below prior to opening. This vial contains a relatively low volume of reagent (25 µL). To minimize loss of volume dilute 1:10 by adding 225 µL of the buffer stated above directly to the vial. Recap, mix thoroughly and briefly centrifuge to collect the volume at the bottom of the vial. Use this intermediate dilution when calculating final dilutions as recommended below. Store the vial at -20°C or below after dilution. Avoid cycles of freezing and thawing.
Dry Ice Shipping	**Please note: This product requires shipment on dry ice. A dry ice surcharge will apply.**
Note	For research use only
NCBI	**31543500**
Expiration Date	12 months from date of receipt.

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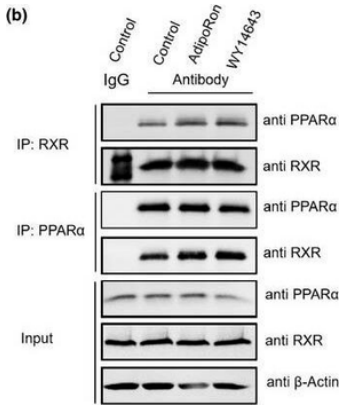
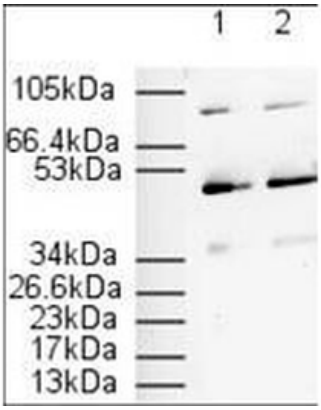


Adipopnectin regulates the expression of ADAM10 and Notch1 through PPARα and JNK pathway respectively. (a) Representative immunoblots and quantification of hippocampal protein levels after chronic ICV injection of AdipoRon, WY14643 (PPARα agonist) and GW6471 (PPARα antagonist) in 12-month-old mice, including Notch1 and ADAM10. Control, n = 6; AdipoRon, n = 6; WY14643, n = 7; AdipoRon + GW6471, n = 7. *p 0.05, **p 0.01, ***p 0.001 compared with Control; #p 0.05, ###p 0.001 compared with AdipoRon treatment. (b) Co-immunoprecipitation results showing chronic ICV injection of AdipoRon or WY14643 enhances the interaction between PPARα and RXR. Control, n = 6; AdipoRon, n = 6; WY14643, n = 7. ***p 0.001 compared with Control. (c) qPCR results showing chronic ICV injection of AdipoRon or WY14643 Induces the recruitment of RXR to the ADAM10 Promoter. Control, n = 6; AdipoRon, n = 7; WY14643, n = 7. ***p 0.001 compared with Control. (d) Chronic ICV injection of AdipoRon upregulates the activity of JNK. Control, n = 6; AdipoRon, n = 6. ***p 0.001. (e) Representative immunoblots and quantification of hippocampal protein levels after chronic ICV injection of AdipoRon, Vinblastine (JNK agonist) and SP600125 (JNK antagonist) in 12-month-old mice, including Notch1 and ADAM10. Control, n = 6; AdipoRon, n = 7; Vinblastine, n = 7; AdipoRon + SP600125, n = 7. **p 0.01, ***p 0.001 compared with Control; ###p 0.001 compared with AdipoRon treatment. Data are presented as means ± SEM.



Affinity Purified Anti-PPAR alpha (N -terminal specific) (Rabbit) is shown to detect a 52 kDa band corresponding to PPAR alpha present in a 3T3 whole cell lysate. Approximately 20 µg of lysate was loaded per lane for SDS-PAGE. Detection occurred after using a 1:500 (lane 1) or 1:1000 (lane 2) dilution of antibody followed by 1:2000 dilution of HRP Goat-a-Rabbit IgG for visualization.

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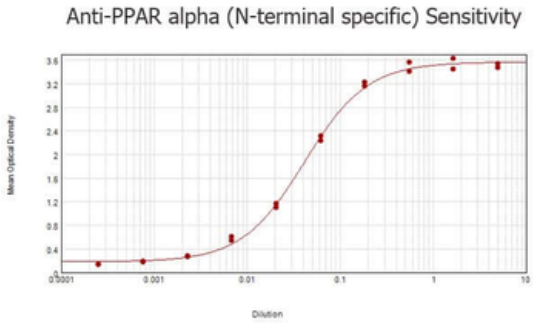
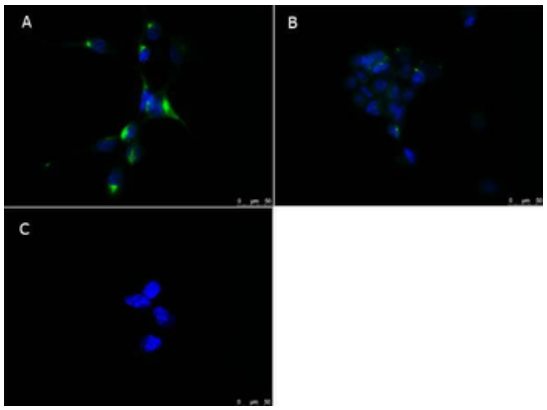
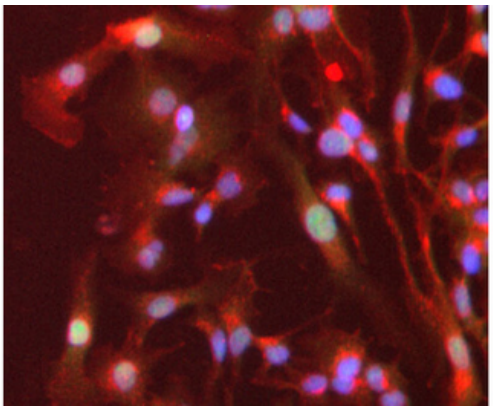


ELISA results of purified Rabbit anti-PPAR Alpha (N-terminal specific) Antibody tested against BSA-conjugated peptide of immunizing peptide. Each well was coated in duplicate with 0.1 µg of conjugate. The starting dilution of antibody was 5 µg/ml and the X-axis represents the Log10 of a 3-fold dilution. This titration is a 4-parameter curve fit where the IC50 is defined as the titer of the antibody. Assay performed using 3% fish gel, Goat anti-Rabbit IgG Antibody Peroxidase Conjugated (Min X Bv Ch Gt GP Ham Hs Hu Ms Rt & Sh Serum Proteins) (p/n orb347654) and TMB ELISA Peroxidase Substrate (p/n orb348651).



Immunofluorescence microscopy of Rabbit Anti-PPAR alpha (N-terminal specific) antibody using (A) Mouse NIH/3T3 or (B) Human HEK293 cells fixed with MeOH. (C) Secondary antibody only with NIH/3T3 cells. Anti-PPAR alpha antibody was used at 10 µg/ml, 1h at RT⁰. Secondary antibody: Anti-RABBIT IgG DyLight™ 488 Conjugated Preadsorbed at 5 ug/ml for 1 h at RT. Staining: PPAR as green fluorescent signal with DAPI (blue) nuclear counterstain.



Immunofluorescence Microscopy of Rabbit anti-PPAR alpha antibody. Tissue: HepG2 cells. Fixation: 4% formaldehyde fixed (10 min). Antigen retrieval: not required. Primary antibody: PPAR alpha antibody at 1 µg/ml overnight at 4°C. Secondary antibody: Alexa Fluor® 488 goat anti-rabbit IgG (H+L) (green) used at a 1:1000, Alexa Fluor® 594 WGA was used to label plasma membranes (red) at a 1:200 dilution for 1h for 45 min at RT. Localization: PPAR alpha is nuclear and occasionally cytoplasmic. Staining: PPAR alpha as green fluorescent signal with DAPI (blue) nuclear counterstain.

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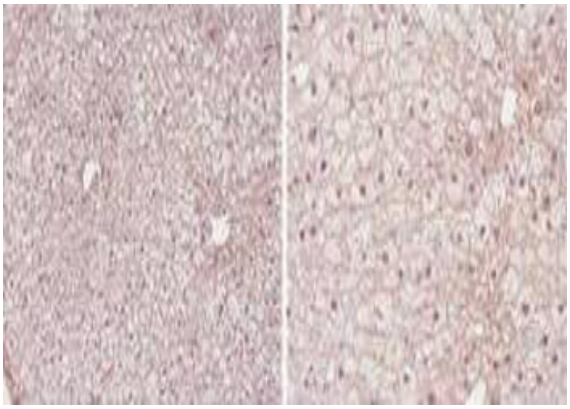
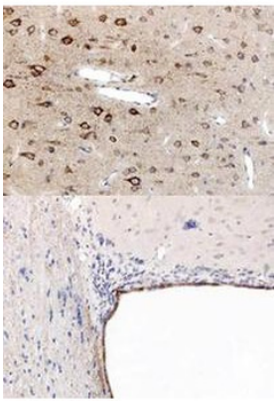
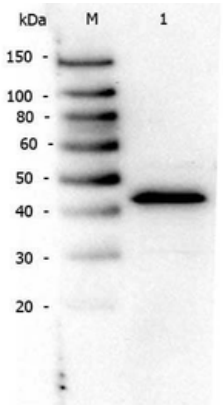


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) showing Biorbyt's PPAR alpha antibody staining of PPAR alpha protein in mouse liver tissue section (Formalin/PFA-fixed paraffin-embedded sections). Tissue underwent formaldehyde fixation before enzymatic antigen retrieval with 0.05% protease in PBS for 5 minutes. Sample was then blocked with 5% serum for 20 minutes at 20°C. The primary antibody was diluted 1:50 and incubated with sample in Tris plus 5% normal goat serum for 1 hour at 20°C. A Biotin conjugated goat polyclonal to rabbit IgG was used at dilution at 1:500 as secondary antibody. Images show nuclear staining in hepatocytes (perfusion-fixed mouse, 10 and 40x microscope magnification).



Immunohistochemistry using Biorbyt's anti-PPAR antibody, showing staining of PPAR alpha in rat brain sections, highlighting cytoplasmic staining in ependymal cells and neurons in frontal cortex. Bottom image shows subventricular zone (svz) of lateral ventrical (exit point of progenitor olfactory neurones); top image shows frontal cortex in the same section. Cytoplasmic staining is also observed in the corpus callosum (bottom image) and in dendritic fields of the cortex. Formalin/PFA-fixed paraffin-embedded sections of rat brain tissue were incubated with the primary antibody at 1:200 for 1 hour. Antigen retrieval was performed by heat induction in citrate buffer pH6.0.



Western Blot of Rabbit anti-PPAR Alpha (N-terminal Specific) antibody. Lane M: Prestained Molecular Weight Markers. Lane 1: NIH/3T3 (p/n orb348714). Load: 10 µg per lane. Primary antibody: PPAR Alpha (N-terminal specific) antibody at 1:1000 for overnight at 4°C. Secondary antibody: Peroxidase rabbit secondary antibody (p/n orb347654) at 1:40000 for 30 min at RT. Block: Blocking Buffer for Fluorescent Western Blotting (p/n orb348637) at RT for 30 min. Predicted/Observed size: ~50 kDa for PPAR Alpha.

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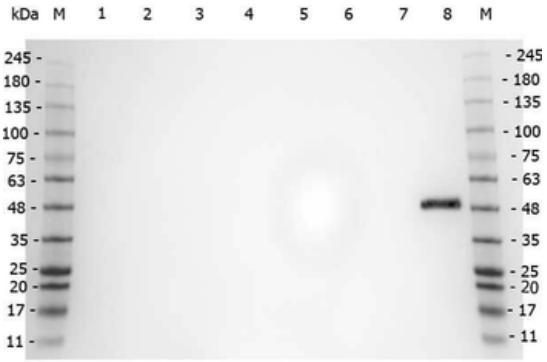

Western Blot of Rabbit anti-PPAR Alpha (N-terminal specific) antibody. Marker: Opal Pre-stained ladder. Lane 1: HEK293 lysate (p/n orb348669). Lane 2: HeLa Lysate (p/n orb348668). Lane 3: MCF-7 Lysate (p/n orb348664). Lane 4: Jurkat Lysate. Lane 5: A431 Lysate (p/n orb348665). Lane 6: LNCaP Lysate (p/n orb348694). Lane 7: A-172 Lysate (p/n orb348708). Lane 8: NIH/3T3 Lysate (p/n orb348714). Load: 35 µg per lane. Primary antibody: PPAR Alpha (N-terminal specific) antibody at 1 ug/ml overnight at 4°C. Secondary antibody: Peroxidase rabbit secondary antibody (p/n orb347654) at 1:30000 for 60 min at RT. Blocking Buffer: 1% Casein-TTBS for 30 min at RT. Predicted/Observed size: 52 kDa for PPAR Alpha.

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