Anti-Apolipoprotein E Antibody [SC0536] - BSA and Azide free HA750206

Product Type:	Recombinant Rabbit monoclonal IgG, primary antibodies
Species reactivity:	Human, Rat
Applications:	WB, IF-Cell, IF-Tissue, IHC-P, IP
Molecular Wt:	Predicted band size: 36 kDa
Clone number:	SC0536
Description:	Apolipoprotein-E (apoE) is a protein component of plasma lipoproteins that mediates the binding, internalization and catabolism of lipoprotein particles. It can serve as a ligand for several lipoprotein receptors, including the LDL (ApoB/E) receptor and the hepatic apoE (chylomicron remnant) receptor. apoE is produced in most organs and occurs in all plasma lipoprotein fractions, constituting 10-20% of VLDL (very low density lipoprotein) and 1-2% of HDL (high density lipoprotein). Three major isoforms of apoE have been described in human (E2, E3 and E4) which differ by only one or two amino acids. Estrogen receptor has been shown to upregulate apoE gene expression via the ERa-mediated pathway, indicating a potential role for apoE in atherosclerosis. This is consistent with studies in mice in which plasma apoE levels were raised, thereby protecting the mice from diet-induced atherosclerosis. apoE has also been shown to be a potent inhibitor of proliferation and thus may play a role in angiogenesis, tumor cell growth and metastasis.
lmmunogen:	Synthetic peptide within C-terminal human Apolipoprotein E.
Positive control:	Human plasma tissue lysates, human tonsil tissue, human liver tissue, human kidney tissue, human placenta tissue, human liver tissue, rat liver, HepG2.
Subcellular location:	Secreted, extracellular space, extracellular matrix.
Database links:	SwissProt: P02649 Human P02650 Rat
Recommended Dilutions: WB IF-Cell IF-Tissue IHC-P IP	1:1,000-1:5,000 1:50-1:200 1:50-1:200 1:50-1:1,000 Use at an assay dependent concentration.
Storage Buffer:	PBS (pH7.4).
Storage Instruction:	Store at +4 $^{\circ}$ C after thawing. Aliquot store at -20 $^{\circ}$ C or -80 $^{\circ}$ C. Avoid repeated freeze / thaw cycles.
	cycles.

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11.

HA750206 - Page 2

Images

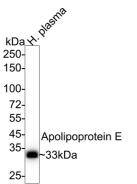


Fig1: Western blot analysis of Apolipoprotein E on human plasma tissue lysates with Rabbit anti-Apolipoprotein E antibody (HA750206) at 1/1,000 dilution.

Lysates/proteins at 20 µg/Lane.

Predicted band size: 36 kDa Observed band size: 33 kDa

Exposure time: 6 seconds;

4-20% SDS-PAGE gel.

Proteins were transferred to a PVDF membrane and blocked with 5% NFDM/TBST for 1 hour at room temperature. The primary antibody (HA750206) at 1/1,000 dilution was used in 5% NFDM/TBST at 4° C overnight. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 1/50,000 dilution was used for 1 hour at room temperature.

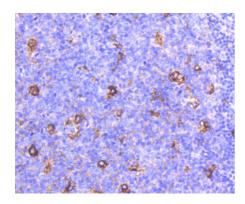


Fig2: Immunohistochemical analysis of paraffin-embedded human tonsil tissue using anti-Apolipoprotein E antibody. The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 8.0-8.4) for 20 minutes. The tissues were blocked in 5% BSA for 30 minutes at room temperature, washed with ddH₂O and PBS, and then probed with the primary antibody (HA750206, 1/50) for 30 minutes at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.

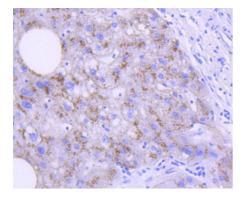


Fig3: Immunohistochemical analysis of paraffin-embedded human liver tissue using anti-Apolipoprotein E antibody. The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 8.0-8.4) for 20 minutes. The tissues were blocked in 5% BSA for 30 minutes at room temperature, washed with ddH₂O and PBS, and then probed with the primary antibody (HA750206, 1/50) for 30 minutes at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.

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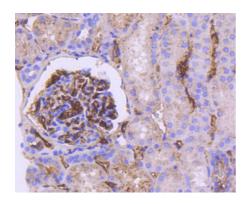


Fig4: Immunohistochemical analysis of paraffin-embedded human kidney tissue using anti-Apolipoprotein E antibody. The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 8.0-8.4) for 20 minutes. The tissues were blocked in 5% BSA for 30 minutes at room temperature, washed with ddH₂O and PBS, and then probed with the primary antibody (HA750206, 1/50) for 30 minutes at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.

Fig5: Immunohistochemical analysis of paraffin-embedded human placenta tissue with Rabbit anti-Apolipoprotein E antibody (HA750206) at 1/1,000 dilution.

The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 20 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH₂O and PBS, and then probed with the primary antibody (HA750206) at 1/1,000 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.

Fig6: Western blot analysis of Apolipoprotein E on different lysates with Rabbit anti-Apolipoprotein E antibody (HA750206) at 1/2,000 dilution.

Lane 1: HepG2 cell lysate Lane 2: Human liver tissue lysate Lane 3: Human plasma tissue lysate

Lysates/proteins at 20 µg/Lane1 and 40ug/Lane2-3.

Predicted band size: 35 kDa Observed band size: 35 kDa

Exposure time: 20 seconds; ECL: K1801;

4-20% SDS-PAGE gel.

Proteins were transferred to a PVDF membrane and blocked with 5% NFDM/TBST for 1 hour at room temperature. The primary antibody (HA750206) at 1/2,000 dilution was used in 5% NFDM/TBST at 4° C overnight. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 1/50,000 dilution was used for 1 hour at room temperature.

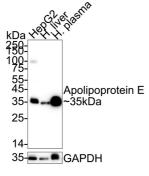
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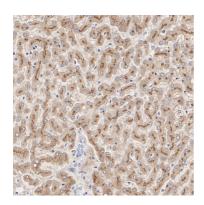


Fig7: Immunohistochemical analysis of paraffin-embedded human liver tissue with Rabbit anti-Apolipoprotein E antibody (HA750206) at 1/1,000 dilution.

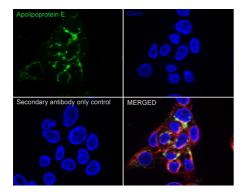
The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 20 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH₂O and PBS, and then probed with the primary antibody (HA750206) at 1/1,000 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.



Fig8: Immunohistochemical analysis of paraffin-embedded rat liver tissue with Rabbit anti-Apolipoprotein E antibody (HA750206) at 1/1,000 dilution.

The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 20 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH₂O and PBS, and then probed with the primary antibody (HA750206) at 1/1,000 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.

Fig9: Immunocytochemistry analysis of HepG2 cells labeling Apolipoprotein E with Rabbit anti-Apolipoprotein E antibody (HA750206) at 1/100 dilution.



Cells were fixed in 4% paraformaldehyde for 15 minutes at room temperature, permeabilized with 0.1% Triton X-100 in PBS for 15 minutes at room temperature, then blocked with 1% BSA in 10% negative goat serum for 1 hour at room temperature. Cells were then incubated with Rabbit anti-Apolipoprotein E antibody (HA750206) at 1/100 dilution in 1% BSA in PBST overnight at 4 °C. Goat Anti-Rabbit IgG H&L (iFluor™ 488, HA1121) was used as the secondary antibody at 1/1,000 dilution. PBS instead of the primary antibody was used as the secondary antibody only control. Nuclear DNA was labelled in blue with DAPI.

Beta tubulin (M1305-2, red) was stained at 1/100 dilution overnight at $+4^{\circ}$ C. Goat Anti-Mouse IgG H&L (iFluor 1594, HA1126) was used as the secondary antibody at 1/1,000 dilution.

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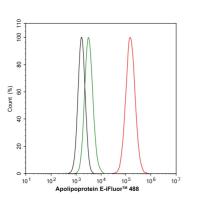


Fig10: Flow cytometric analysis of HepG2 cells labeling Apolipoprotein E.

Cells were fixed and permeabilized. Then stained with the primary antibody (HA750206, 1µg/mL) (red) compared with Rabbit IgG Isotype Control (green). After incubation of the primary antibody at +4°C for an hour, the cells were stained with a iFluor[™] 488 conjugate-Goat anti-Rabbit IgG Secondary antibody (HA1121) at 1/1,000 dilution for 30 minutes at +4°C. Unlabelled sample was used as a control (cells without incubation with primary antibody; black).

Note: All products are "FOR RESEARCH USE ONLY AND ARE NOT INTENDED FOR DIAGNOSTIC OR THERAPEUTIC USE".

Background References

- 1. Yang LG et al. Apolipoprotein E in lipid metabolism and neurodegenerative disease. Trends Endocrinol Metab. 2023 Aug
- 2. Saito T et al. Apolipoprotein E-related glomerular disorders. Kidney Int. 2020 Feb

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Applications:WB=Western blot IHC-P=Immunohistochemistry (paraffin) IF-Cell=Immunofluorescence (Cell) IF-Tissue=Immunofluorescence (Tissue) FC=Flow cytometry IP=Immunoprecipitation

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