Overview

Product name: Anti-HNF-4-alpha antibody [EPR3648]
Description: Rabbit monoclonal [EPR3648] to HNF-4-alpha
Host species: Rabbit
Tested applications: Suitable for: WB, IHC-P, Flow Cyt, ICC/IF
Unsuitable for: IP
Species reactivity: Reacts with: Human
Immunogen: Synthetic peptide within Human HNF-4-alpha aa 1-100. The exact sequence is proprietary.
General notes: A trial size is available to purchase for this antibody.
Mouse, Rat: We have preliminary internal testing data to indicate this antibody may not react with these species. Please contact us for more information.
Our RabMAb® technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to RabMab® patents

We are constantly working hard to ensure we provide our customers with best in class antibodies. As a result of this work we are pleased to now offer this antibody in purified format. We are in the process of updating our datasheets. The purified format is designated 'PUR' on our product labels. If you have any questions regarding this update, please contact our Scientific Support team.

This product is a recombinant rabbit monoclonal antibody.

Properties

Form: Liquid
Storage buffer: pH: 7.20
Preservative: 0.01% Sodium azide
Constituents: 59% PBS, 40% Glycerol, 0.05% BSA
Purity: Protein A purified
Clonality: Monoclonal
Clone number: EPR3648
Isotype: IgG

Applications

Our Abpromise guarantee covers the use of ab92378 in the following tested applications.

The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

<table>
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<tr>
<th>Application</th>
<th>Abreviews</th>
<th>Notes</th>
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<tbody>
<tr>
<td>IHC-P</td>
<td>⭐⭐⭐⭐⭐</td>
<td>1/250 - 1/500. Perform heat mediated antigen retrieval before commencing with IHC staining protocol. See IHC antigen retrieval protocols. For unpurified use at 1/100 - 1/250.</td>
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<tr>
<td>Flow Cyt</td>
<td></td>
<td>1/70. For unpurified use at 1/100. ab172730 - Rabbit monoclonal IgG, is suitable for use as an isotype control with this antibody.</td>
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<tr>
<td>ICC/IF</td>
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<td>1/100 - 1/250.</td>
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Application notes: Is unsuitable for IP.

Target

Function: Transcriptionally controlled transcription factor. Binds to DNA sites required for the transcription of alpha 1-antitrypsin, apolipoprotein CIII, transthyretin genes and HNF1-alpha. May be essential for development of the liver, kidney and intestine.

Involvement in disease: Defects in HNF4A are the cause of maturity-onset diabetes of the young type 1 (MODY1) [MIM:125850]; also symbolized MODY-1. MODY is a form of diabetes that is characterized by an autosomal dominant mode of inheritance, onset in childhood or early adulthood (usually before 25 years of age), a primary defect in insulin secretion and frequent insulin-independence at the beginning of the disease.

Sequence similarities: Belongs to the nuclear hormone receptor family. NR2 subfamily. Contains 1 nuclear receptor DNA-binding domain.

Post-translational modifications: Phosphorylated on tyrosine residue(s); phosphorylation is important for its DNA-binding activity. Phosphorylation may directly or indirectly play a regulatory role in the subnuclear distribution.

Cellular localization: Nucleus.

Images
Anti-HNF-4-alpha antibody [EPR3648] (ab92378) at 1/2000 dilution (purified) + SW480 cell lysate at 20 µg

Secondary
Peroxidase conjugated goat anti-rabbit IgG (H+L) at 1/1000 dilution

**Predicted band size:** 53 kDa
**Observed band size:** 53 kDa

Blocking and dilution buffer: 5% NFDM/TBST.

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human liver tissue labelling HNF-4-alpha with purified ab92378 at 1/100. Heat mediated antigen retrieval was performed using Tris/EDTA buffer pH 9. **ab97051**, a HRP-conjugated goat anti-rabbit IgG (H+L) was used as the secondary antibody (1/500). Negative control using PBS instead of primary antibody. Counterstained with hematoxylin.
Immunocytochemistry/Immunofluorescence analysis of HepG2 cells labelling HNF-4 with purified ab92378 at 1/100. Cells were fixed with 4% paraformaldehyde and permeabilized with 0.1% Triton X-100. ab150077, an Alexa Fluor® 488-conjugated goat anti-rabbit IgG (1/500) was used as the secondary antibody. DAPI (blue) was used as the nuclear counterstain. ab7291, a mouse anti-tubulin (1/500) and ab150120, an Alexa Fluor® 594-conjugated goat anti-mouse IgG (1/500) were also used.

Control 1: primary antibody (1/100) and secondary antibody, ab150120, an Alexa Fluor® 594-conjugated goat anti-mouse IgG (1/500).
Control 2: ab7291 (1/1000) and secondary antibody, ab150077, an Alexa Fluor® 488-conjugated goat anti-rabbit IgG (1/500).

Anti-HNF-4-alpha antibody [EPR3648] (ab92378) at 1/5000 dilution (purified) + HepG2 cell lysate at 10 µg

**Secondary**
Peroxidase conjugated goat anti-rabbit IgG (H+L) at 1/1000 dilution

**Predicted band size:** 53 kDa

**Observed band size:** 53 kDa

Blocking and dilution buffer: 5% NFDM/TBST.

**All lanes** : Anti-HNF-4-alpha antibody [EPR3648] (ab92378) at 1/1000 dilution (unpurified)

**Lane 1** : HepG2 cell lysate
**Lane 2** : A549 cell lysate
**Lane 3** : SW480 cell lysate

Lysates/proteins at 10 µg per lane.

**Secondary**
**All lanes** : HRP-conjugated goat anti-rabbit IgG at 1/2000 dilution

**Predicted band size:** 53 kDa
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human colon tissue (A) and human kidney tissue (B) labelling HNF-4-alpha with unpurified ab92378 at a 1/100 dilution. Detection: DAB staining.

Immunocytochemistry/Immunofluorescence analysis of HepG2 cells labelling HNF-4-alpha with unpurified ab92378 at a 1/100 dilution.

Flow Cytometry analysis of HepG2 cells labelling HNF-4 with purified ab92378 at 1/70 (red). Cells were fixed with 2% paraformaldehyde. A FITC-conjugated goat anti-rabbit IgG (1/150) was used as the secondary antibody. Black - Isotype control, rabbit monoclonal IgG. Blue - Unlabelled control, cells without incubation with primary and secondary antibodies.
Overlay histogram showing HepG2 cells stained with unpurified ab92378 (red line). The cells were fixed with 80% methanol (5 min) and then permeabilized with 0.1% PBS-Tween for 20 min. The cells were then incubated in 1x PBS / 10% normal goat serum / 0.3M glycine to block non-specific protein-protein interactions. The cells were then incubated with the antibody (unpurified ab92378, 1/100 dilution) for 30 min at 22ºC. The secondary antibody used was DyLight® 488 goat anti-rabbit IgG (H+L) (ab96899) at 1/500 dilution for 30 min at 22ºC. Isotype control antibody (black line) was rabbit IgG (monoclonal) (1µg/1x10^6 cells) used under the same conditions. Acquisition of >5,000 events was performed. This antibody gave a positive signal in HepG2 cells fixed with 4% paraformaldehyde (10 min)/permeabilized with 0.1% PBS-Tween for 20 min used under the same conditions.

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

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