# abcam

# Product datasheet

# Anti-NQO1 antibody [A180] ab28947





★★★★★ 12 Abreviews 132 References 10 Images

#### Overview

**Product name** Anti-NQO1 antibody [A180]

**Description** Mouse monoclonal [A180] to NQO1

**Host species** Mouse

**Tested applications** Suitable for: Sandwich ELISA, ICC/IF, WB, IHC-P, Flow Cyt (Intra)

Species reactivity Reacts with: Human

Predicted to work with: Mouse, Rat, Dog, Monkey

**Immunogen** Recombinant full length protein corresponding to Human NQO1.

Positive control WB: HAP1 and HepG2 whole cell lysates; human kidney tissue lysate. ICC/IF: HepG2 cells. Flow

Cyt (Intra): HeLa cells. IHC-P: FFPE human breast adenocarcinoma and pancreas

adenocarcinoma tissue sections.

**General notes** This antibody clone is manufactured by Abcam. If you require a custom buffer formulation or

conjugation for your experiments, please contact orders@abcam.com.

The Life Science industry has been in the grips of a reproducibility crisis for a number of years. Abcam is leading the way in addressing this with our range of recombinant monoclonal antibodies and knockout edited cell lines for gold-standard validation. Please check that this product meets

your needs before purchasing.

If you have any questions, special requirements or concerns, please send us an inquiry and/or contact our Support team ahead of purchase. Recommended alternatives for this product can be

found below, along with publications, customer reviews and Q&As

#### **Properties**

**Form** Liquid

Storage instructions Shipped at 4°C. Upon delivery aliquot. Store at -20°C or -80°C. Avoid freeze / thaw cycle.

Storage buffer

Preservative: 0.02% Sodium azide

Constituent: PBS

Some batches contain 6.97% L-Arginine as a stabilizing agent. For lot-specific buffer information,

please contact our Scientific Support team.

**Purity** Protein G purified

**Clonality** Monoclonal

Clone number A180 Isotype IgG1

# **Applications**

The Abpromise guarantee Our Abpromise guarantee covers the use of ab28947 in the following tested applications.

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

Application	Abreviews	Notes
Sandwich ELISA		1/500. Can be used as capture antibody in conjunction with <u>ab34173</u> as detection antibody.
ICC/IF	<b>★★★★★ (2)</b>	Use a concentration of 5 µg/ml.
WB	★ ★ ★ 🖨 🛣 (4)	Use a concentration of 1 µg/ml. Predicted molecular weight: 30 kDa.
IHC-P	<b>★★★★★ (4)</b>	Use at an assay dependent concentration.
Flow Cyt (Intra)		Use 1µg for 10 <sup>6</sup> cells.  ab170190 - Mouse monoclonal lgG1, is suitable for use as an isotype control with this antibody.

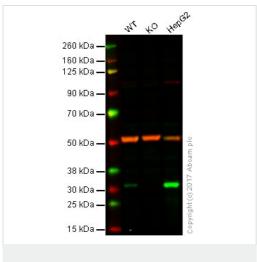
Function	The enzyme apparently serves as a quinone reductase in connection with conjugation reaction	
	hydroquinons involved in detoxification pathways as well as in biosynthetic processes such as the	
	vitamin K-dependent gamma-carboxylation of glutamate residues in prothrombin synthesis.	
Sequence similarities	Belongs to the NAD(P)H dehydrogenase (quinone) family.	

belongs to the W.B.(1) in denigning ended (quinone) raminy.

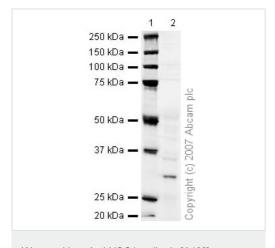
Cellular localization Cytoplasm.

# **Images**

**Target** 



Western blot - Anti-NQO1 antibody [A180] (ab28947)



Western blot - Anti-NQO1 antibody [A180] (ab28947)

Lane 1: Wild-type HAP1 whole cell lysate (20 µg)

Lane 2: NQO1 knockout HAP1 whole cell lysate (20 µg)

Lane 3: HepG2 whole cell lysate (20 µg)

**Lanes 1 - 3:** Merged signal (red and green). Green - ab28947 observed at 31 kDa. Red - loading control, **ab176560**, observed at 50 kDa.

ab28947 was shown to specifically react with NQO1 in wild-type HAP1 cells as signal was lost in NQO1 knockout cells. Wild-type and NQO1 knockout samples were subjected to SDS-PAGE. ab28947 and <a href="mailto:ab176560">ab176560</a> (Rabbit anti-alpha Tubulin loading control) were incubated overnight at 4°C at 1 µg/ml and 1/20000 dilution respectively. Blots were developed with Goat anti-Mouse lgG H&L (IRDye® 800CW) preabsorbed <a href="mailto:ab216772">ab216772</a> and Goat anti-Rabbit lgG H&L (IRDye® 680RD) preabsorbed <a href="mailto:ab216777">ab216777</a> secondary antibodies at 1/20000 dilution for 1 hour at room temperature before imaging.

Lane 1: Marker

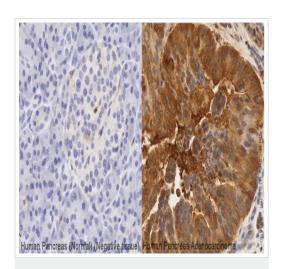
Lane 2: Anti-NQO1 antibody [A180] (ab28947) at 1 µg/ml

Lane 2: Kidney (Human) Tissue Lysate (ab7920) at 20 µg

#### Secondary

Lane 2: IRDye 680 Conjugated Goat Anti-Mouse IgG (H+L) at 1/10000 dilution

Predicted band size: 30 kDa
Observed band size: 30 kDa

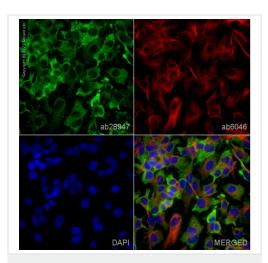


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-NQO1 antibody [A180] (ab28947)

IHC image of NQO1 staining in sections of formalin fixed paraffin embedded normal human pancreas\* (left) and human pancreas adenocarcinoma\* (right), performed on a Leica BOND<sup>TM</sup> system using the standard protocol F. The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH6, epitope retrieval solution 1) for 20 mins. The section was then incubated with ab28947, 0.1µg/ml, for 15 mins at room temperature and detected using an HRP conjugated compact polymer system. DAB was used as the chromogen. The section was then counterstained with haematoxylin and mounted with DPX.

For other IHC staining systems (automated and non-automated) customers should optimize variable parameters such as antigen retrieval conditions, primary antibody concentration and antibody incubation times.

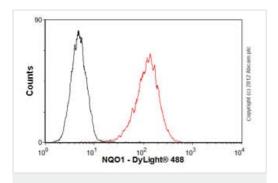
\*Tissue obtained from the Human Research Tissue Bank, supported by the NIHR Cambridge Biomedical Research Centre



Immunocytochemistry/ Immunofluorescence - Anti-NQO1 antibody [A180] (ab28947)

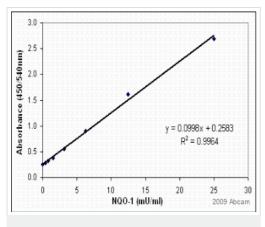
ab28947 staining NQO1 in HepG2 cells. The cells were fixed with 100% methanol (5 min), permeabilized with 0.1% PBS-Tween for 5 minutes and then blocked with 1% BSA/10% normal goat serum/0.3M glycine in 0.1% PBS-Tween for 1h. The cells were then incubated overnight at 4°C with ab28947 at 5 µg/ml and ab6046, Rabbit polyclonal to beta Tubulin - Loading Control. Cells were then incubated with ab150117, Goat polyclonal Secondary Antibody to Mouse IgG H&L (Alexa Fluor® 488) preadsorbed at 1/1000 dilution (shown in green) and ab150080, Goat polyclonal Secondary Antibody to Rabbit IgG - H&L (Alexa Fluor® 594) at 1/1000 dilution (shown in pseudocolour red). Nuclear DNA was labelled with DAPI (shown in blue).

Image was acquired with a high-content analyser (Operetta CLS, Perkin Elmer) and a maximum intensity projection of confocal sections is shown.



Flow Cytometry (Intracellular) - Anti-NQO1 antibody [A180] (ab28947)

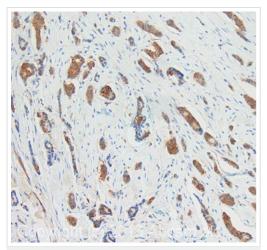
Overlay histogram showing HeLa cells stained with ab28947 (red line). The cells were fixed with 80% methanol (5 min) and then permeabilized with 0.1% PBS-Tween for 20 min. The cells were then incubated in 1x PBS / 10% normal goat serum / 0.3M glycine to block non-specific protein-protein interactions followed by the antibody (ab28947, 1µg/1x10<sup>6</sup> cells) for 30 min at 22°C. The secondary antibody used was goat **anti-mouse DyLight® 488** (lgG H+L) (**ab96879**) at 1/500 dilution for 30 min at 22°C. Isotype control antibody (black line) was mouse lgG1 [ICIGG1] (**ab91353**, 2µg/1x10<sup>6</sup> cells) used under the same conditions. Acquisition of >5,000 events was performed. This antibody gave a positive signal in HeLa cells fixed with 4% paraformaldehyde (10 min)/permeabilized with 0.1% PBS-Tween for 20 min used under the same conditions.



Sandwich ELISA - Anti-NQO1 antibody [A180] (ab28947)

This image is courtesy of an anonymous Abreview

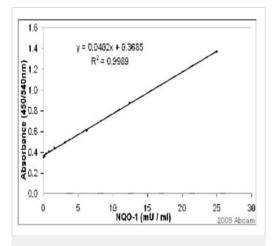
Sandwich ELISA for the detection of NQO1. ab28947 (1/500) was used as the capture antibody. A rabbit polyclonal raised againts the C-terminal end of NQO1 was used for the detection. Please refer to abreview for further experimental details.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-NQO1 antibody [A180] (ab28947)

IHC image of NQO1 staining in human breast adenocarcinoma formalin fixed paraffin embedded tissue section, performed on a Leica Bond system using the standard protocol F. The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH6, epitope retrieval solution 1) for 20 mins. The section was then incubated with ab28947, 0.1µg/ml, for 15 mins at room temperature and detected using an HRP conjugated compact polymer system. DAB was used as the chromogen. The section was then counterstained with haematoxylin and mounted with DPX.

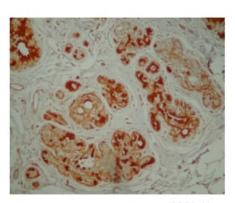
For other IHC staining systems (automated and non-automated) customers should optimize variable parameters such as antigen retrieval conditions, primary antibody concentration and antibody incubation times.



Sandwich ELISA - Anti-NQO1 antibody [A180] (ab28947)

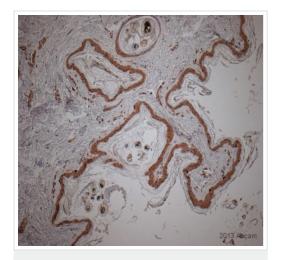
Sandwich ELISA for the detection of NQO1, using ab28947 (1/500) as the capture antibody and <u>ab34173</u> (1/1000) for the detection.

Human breast cancer tissue stained with ab28947 NQO1 antibody.



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Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-NQO1 antibody [A180] (ab28947)



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-NQO1 antibody [A180] (ab28947)

This image is courtesy of an anonymous Abreview

Immunohistochemical analysis of dog skin tissue, staining NQO1 with ab28947.

Tissue was fixed with formaldehyde and antigen retrieval was by heat mediation in a citrate buffer (pH 6). Samples were incubated with primary antibody (1/175 in BSA in TBS) for 45 minutes.

ab98784 rabbit polyclonal to anti-mouse HRP (lgG (1/500) was used as the secondary antibody.

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