abcam

Product datasheet

Anti-ACE2 antibody [EPR4435(2)] - BSA and Azide free ab239924

KO VALIDATED Recombinant RabMAb

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Overview

Product name	Anti-ACE2 antibody [EPR4435(2)] - BSA and Azide free
Description	Rabbit monoclonal [EPR4435(2)] to ACE2 - BSA and Azide free
Host species	Rabbit
Tested applications	**Suitable for:** Indirect ELISA, IP, IHC-P, WB **Unsuitable for:** Flow Cyt or ICC/IF
Species reactivity	**Reacts with:** Mouse, Rat, Human
Immunogen	Synthetic peptide. This information is proprietary to Abcam and/or its suppliers. (Peptide available as **ab198988**)
Positive control	IHC-P: Human, mouse, and rat kidney tissues; WB: Caco-2, HepG2 and Calu-3 cell lysates; Human fetal kidney and human testis lysates; Human and rat heart tissue lysate; Human lung tissue lysate; Mouse and rat spleen, testis lung tissue lysate; IP: Human testis tissue lysate.
General notes	ab239924 is the carrier-free version of **ab108252**. Our **carrier-free** antibodies are typically supplied in a PBS-only formulation, purified and free of BSA, sodium azide and glycerol. The carrier-free buffer and high concentration allow for increased conjugation efficiency. This conjugation-ready format is designed for use with fluorochromes, metal isotopes, oligonucleotides, and enzymes, which makes them ideal for antibody labelling, functional and cell-based assays, flow-based assays (e.g. mass cytometry) and Multiplex Imaging applications. Use our **conjugation kits** for antibody conjugates that are ready-to-use in as little as 20 minutes with <1 minute hands-on-time and 100% antibody recovery: available for fluorescent dyes, HRP, biotin and gold. This product is compatible with the Maxpar® Antibody Labeling Kit from Fluidigm, without the need for antibody preparation. Maxpar® is a trademark of Fluidigm Canada Inc. This product is a recombinant monoclonal antibody, which offers several advantages including: - High batch-to-batch consistency and reproducibility - Improved sensitivity and specificity - Long-term security of supply - Animal-free production For more information **see here**.

Our RabMAb® technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to **RabMAb® patents**.

Properties

Form	Liquid
Storage instructions	Shipped at 4°C. Store at +4°C. Do Not Freeze.
Storage buffer	pH: 7.2 Constituent: PBS
Carrier free	Yes
Purity	Protein A purified
Clonality	Monoclonal
Clone number	EPR4435(2)
Isotype	IgG

Applications

The Abpromise guarantee Our **Abpromise guarantee** covers the use of ab239924 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
Indirect ELISA		Use at an assay dependent concentration.
IP		Use at an assay dependent concentration.
IHC-P		Use at an assay dependent concentration. Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.
WB	★★★★☆ **(1)**	Use at an assay dependent concentration. Predicted molecular weight: 92 kDa.Can be blocked with **ACE2 peptide (ab198988)**.

Application notes Is unsuitable for Flow Cyt or ICC/IF.

Target

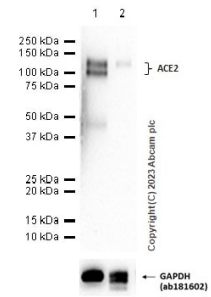
Function Carboxypeptidase which converts angiotensin I to angiotensin 1-9, a peptide of unknown function, and angiotensin II to angiotensin 1-7, a vasodilator. Also able to hydrolyze apelin-13 and dynorphin-13 with high efficiency. May be an important regulator of heart function. In case of human coronaviruses SARS and HCoV-NL63 infections, serve as functional receptor for the spike glycoprotein of both coronaviruses.

Tissue specificity Expressed in endothelial cells from small and large arteries, and in arterial smooth muscle cells. Expressed in lung alveolar epithelial cells, enterocytes of the small intestine, Leydig cells and Sertoli cells (at protein level). Expressed in heart, kidney, testis, and gastrointestinal system.

Sequence similarities Belongs to the peptidase M2 family.

Post-translational modifications	N-glycosylation on Asn-90 may limit SARS infectivity.
Cellular localization	Secreted and Cell membrane.

Images



Western blot - Anti-ACE2 antibody [EPR4435(2)] - BSA and Azide free (ab239924)

All lanes : Anti-ACE2 antibody [EPR4435(2)] (**ab108252**) at 1/1000 dilution

Lane 1 : Human heart tissue lysate
Lane 2 : Rat heart tissue lysate

Lysates/proteins at 20 µg per lane.

Secondary
All lanes : Anti-Rabbit IgG (HRP) with minimal cross-reactivity with human IgG at 1/2000 dilution

Predicted band size: 92 kDa
Observed band size: 110,120 kDa

Exposure time: 180 seconds

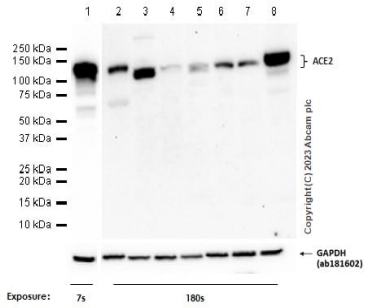
This data was developed using **ab108252**, the same antibody clone in a different buffer formulation.

Blocking and diluting buffer and concentration: 5% NFDM/TBST.

ab181602 was used as a GAPDH loading control.

Two bands observed by **ab108252** corresponding to glycosylation and non-glycosylation forms.

Signal in heart tissue is low, we recommend loading more amount of lysate or using lower antibody dilution to improve result.



Western blot - Anti-ACE2 antibody [EPR4435(2)] - BSA and Azide free (ab239924)

All lanes : Anti-ACE2 antibody [EPR4435(2)] (**ab108252**) at 1/1000 dilution

Lane 1 : Human testis tissue lysate at 20 µg
Lane 2 : Human lung tissue lysate at 20 µg
Lane 3 : Mouse testis tissue lysate
Lane 4 : Mouse spleen tissue lysate
Lane 5 : Mouse lung tissue lysate
Lane 6 : Rat testis tissue lysate
Lane 7 : Rat spleen tissue lysate
Lane 8 : Rat lung tissue lysate

Secondary
All lanes : Anti-Rabbit IgG (HRP) with minimal cross-reactivity with human IgG at 1/2000 dilution

Predicted band size: 92 kDa
Observed band size: 110,120 kDa

This data was developed using **ab108252**, the same antibody clone in a different buffer formulation.

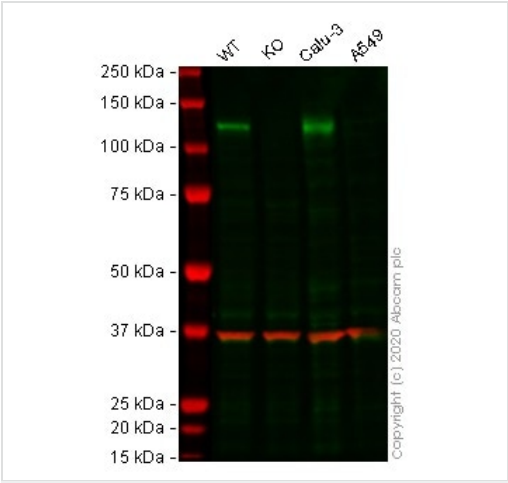
Blocking and diluting buffer and concentration: 5% NFDM/TBST.

ab181602 was used as GAPDH loading control.

Exposure time: Lane 1: 7 seconds; Lane 2-8: 180 seconds.

Two bands observed by **ab108252** corresponding to glycosylation and non-glycosylation forms.

Signal in mouse and rat tissues are low, we recommend loading more amount of lysate or using lower antibody dilution to improve result.



Western blot - Anti-ACE2 antibody [EPR4435(2)] - BSA and Azide free (ab239924)

All lanes : Anti-ACE2 antibody [EPR4435(2)] (**ab108252**) at 1/1000 dilution

Lane 1 : Wild-type HepG2 cell lysate
Lane 2 : ACE2 knockout HepG2 cell lysate
Lane 3 : Calu-3 cell lysate
Lane 4 : A549 cell lysate

Lysates/proteins at 30 µg per lane.

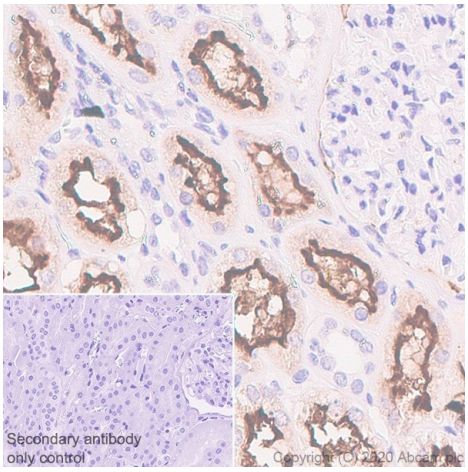
Performed under reducing conditions.

Predicted band size: 92 kDa
Observed band size: 130 kDa

This data was developed using **ab108252**, the same antibody clone in a different buffer formulation.

Lanes 1 - 4: Merged signal (red and green). Green - **ab108252** observed at 130 kDa. Red - loading control **ab8245** (Mouse anti-GAPDH antibody [6C5]) observed at 37kDa.

ab108252 was shown to react with ACE2 in wild-type HepG2 cells in western blot with loss of signal observed in ACE2 knockout cell line **ab273733** (knockout cell lysate **ab275495**). Wild-type and ACE2 knockout HepG2 cell lysates were subjected to SDS-PAGE. Membranes were blocked in 3% milk in TBS-T (0.1% Tween®) before incubation with **ab108252** and **ab8245** (Mouse anti-GAPDH antibody [6C5]) overnight at 4°C at a 1 in 1000 dilution and a 1 in 20000 dilution respectively. Blots were incubated with Goat anti-Rabbit IgG H&L (IRDye® 800CW) preabsorbed (**ab216773**) and Goat anti-Mouse IgG H&L (IRDye® 680RD) preabsorbed (**ab216776**) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.



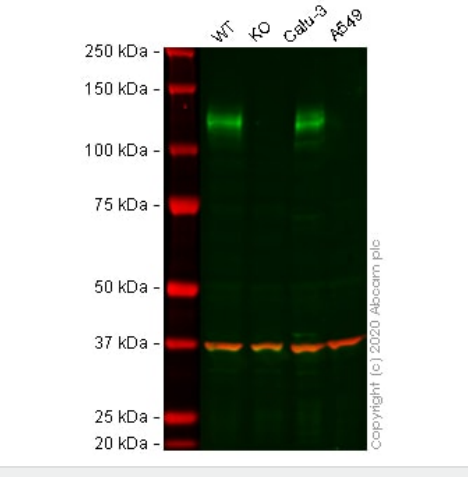
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-ACE2 antibody [EPR4435(2)] - BSA and Azide free (ab239924)

This data was developed using **ab108252**, the same antibody clone in a different buffer formulation.

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human kidney tissue labeling ACE2 with **ab108252** at 1/6400 dilution. Heat mediated antigen retrieval was performed using Bond™ Epitope Retrieval Solution 2 (pH 9.0) for 20 minutes. Staining was visualised using Rabbit specific IHC polymer detection kit HRP/DAB (**ab209101**). Negative control using PBS instead of primary antibody. Counterstained with hematoxylin.

The section was incubated with **ab108252** for 30 mins at room temperature.

The immunostaining was performed on a Leica Biosystems BOND® RX instrument.



Western blot - Anti-ACE2 antibody [EPR4435(2)] - BSA and Azide free (ab239924)

All lanes : Anti-ACE2 antibody [EPR4435(2)] (**ab108252**) at 1/1000 dilution

Lane 1 : Wild-type Caco-2 cell lysate
Lane 2 : ACE2 knockout Caco-2 cell lysate
Lane 3 : Calu-3 cell lysate
Lane 4 : A549 cell lysate

Lysates/proteins at 30 µg per lane.

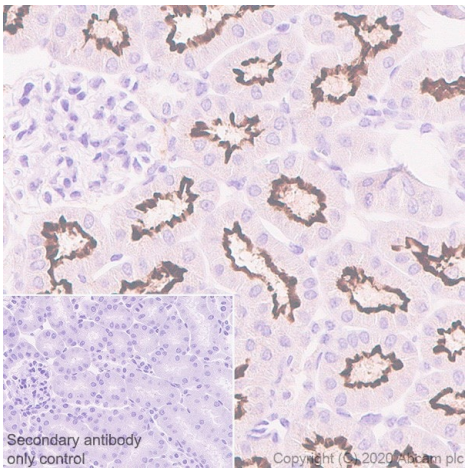
Performed under reducing conditions.

Predicted band size: 92 kDa
Observed band size: 125 kDa

This data was developed using the same antibody clone in a different buffer formulation (**ab108252**).

Lanes 1 - 4: Merged signal (red and green). Green - **ab108252** observed at 125 kDa. Red - loading control **ab8245** (Mouse anti-GAPDH antibody [6C5]) observed at 37kDa.

ab108252 was shown to react with ACE2 in Caco-2 wild-type cells in western blot with loss of signal observed in ACE2 knockout cell line **ab273731** (knockout cell lysate **ab275516**). Wild-type and ACE2 knockout Caco-2 cell lysates were subjected to SDS-PAGE. Membranes were blocked in fluorescent western blot (TBS-based) blocking solution before incubation with **ab108252** and **ab8245** (Mouse anti-GAPDH antibody [6C5]) overnight at 4°C at a 1 in 1000 Dilution and a 1 in 20000 dilution respectively. Blots were incubated with Goat anti-Rabbit IgG H&L (IRDye® 800CW) preabsorbed (**ab216773**) and Goat anti-Mouse IgG H&L (IRDye® 680RD) preabsorbed (**ab216776**) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.



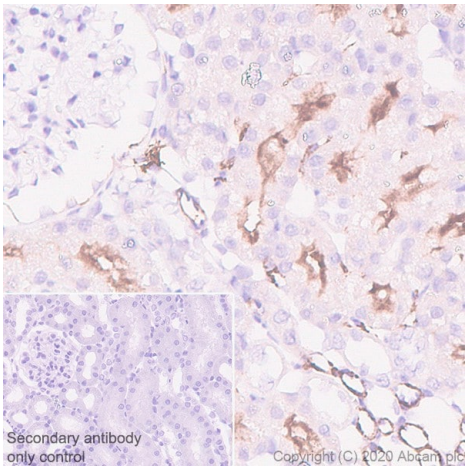
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-ACE2 antibody [EPR4435(2)] - BSA and Azide free (ab239924)

This data was developed using **ab108252**, the same antibody clone in a different buffer formulation.

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of mouse kidney tissue labeling ACE2 with **ab108252** at 1/6400 dilution. Heat mediated antigen retrieval was performed using Bond™ Epitope Retrieval Solution 2 (pH 9.0) for 20 minutes. Staining was visualised using Rabbit specific IHC polymer detection kit HRP/DAB (**ab209101**). Negative control using PBS instead of primary antibody. Counterstained with hematoxylin.

The section was incubated with **ab108252** for 30 mins at room temperature.

The immunostaining was performed on a Leica Biosystems BOND® RX instrument.



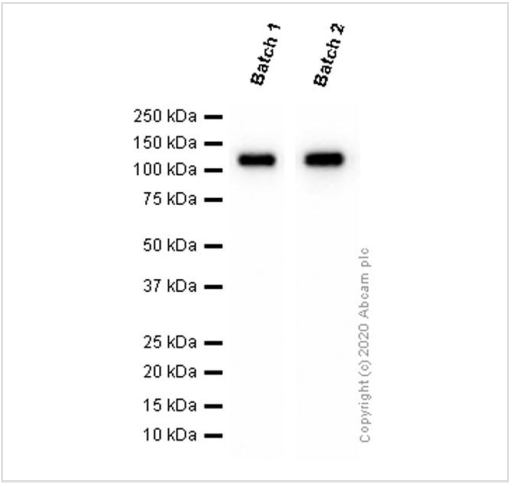
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-ACE2 antibody [EPR4435(2)] - BSA and Azide free (ab239924)

This data was developed using **ab108252**, the same antibody clone in a different buffer formulation.

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of rat kidney tissue labeling ACE2 with **ab108252** at 1/6400 dilution. Heat mediated antigen retrieval was performed using Bond™ Epitope Retrieval Solution 2 (pH 9.0) for 20 minutes. Staining was visualised using Rabbit specific IHC polymer detection kit HRP/DAB (**ab209101**). Negative control using PBS instead of primary antibody. Counterstained with hematoxylin.

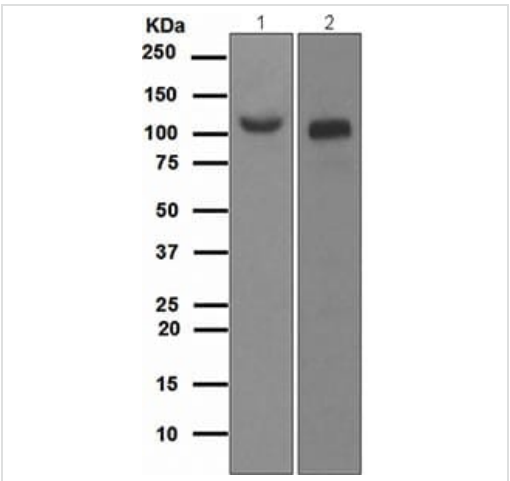
The section was incubated with **ab108252** for 30 mins at room temperature.

The immunostaining was performed on a Leica Biosystems BOND® RX instrument.



Western blot - Anti-ACE2 antibody [EPR4435(2)] - BSA and Azide free (ab239924)

This data was developed using **ab108252**, the same antibody clone in a different buffer formulation. Different batches of **ab108252** were tested on Human kidney lysate at 0.2 µg/ml. 15 µg of lysate was loaded in each lane. Bands observed at 120 kDa.



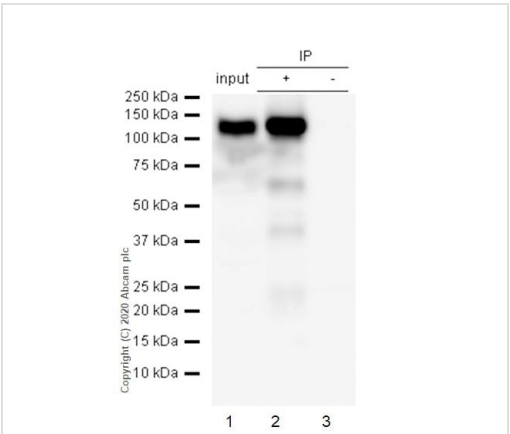
Western blot - Anti-ACE2 antibody [EPR4435(2)] - BSA and Azide free (ab239924)

All lanes : Anti-ACE2 antibody [EPR4435(2)] (**ab108252**) at 1/1000 dilution

Lane 1 : Human fetal kidney lysate
Lane 2 : Human testis lysate

Lysates/proteins at 10 µg per lane.

Predicted band size: 92 kDa



Immunoprecipitation - Anti-ACE2 antibody [EPR4435(2)] - BSA and Azide free (ab239924)

ab239924 Immunoprecipitating ACE2 in human testis tissue lysate. 0.35 mg of tissue lysate was incubated with 2 µg primary antibody (1/50). For western blotting a HRP-conjugated Veriblot for IP Detection Reagent (**ab131366**) (1/1000) was used to confirm successful immunoprecipitation.

Exposure time: 1 second.
Blocking buffer and concentration: 5% NFDM/TBST.
Diluting buffer and concentration: 5% NFDM /TBST.

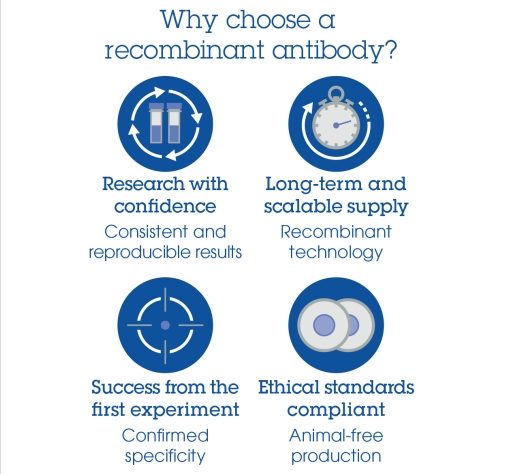
All lanes : Anti-ACE2 antibody [EPR4435(2)] - BSA and Azide free (ab239924) at 1/500 dilution

Lane 1 : Human testis tissue lysate at 10 µg

Lane 2 : ab239924 + Human testis tissue lysate

Lane 3 : Rabbit monoclonal IgG (**ab172730**) instead of ab239924 in Human testis tissue lysate

Observed band size: 110 kDa



Anti-ACE2 antibody [EPR4435(2)] - BSA and Azide free (ab239924)

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