Human/Mouse/Rat/Hamster ACE-2 Antibody

Antigen Affinity-purified Polyclonal Goat IgG
Catalog Number: AF933

DESCRIPTION

Species Reactivity	Human/Mouse/Rat/Hamster
Specificity	Detects human ACE-2 in direct ELISAs. Detects human, mouse, and rat ACE-2 in Western blots. Detects Hamster ACE-2 in immunohistochemistry. In direct ELISAs and Western blots, less than 1% cross-reactivity with recombinant human ACE is observed.
Source	Polyclonal Goat IgG
Purification	Antigen Affinity-purified
Immunogen	Mouse myeloma cell line NS0-derived recombinant human ACE-2 Gln18-Ser740 Accession # Q9BYF1
Formulation	Lyophilized from a 0.2 μm filtered solution in PBS with Trehalose. See Certificate of Analysis for details. *Small pack size (-SP) is supplied either lyophilized or as a 0.2 μm filtered solution in PBS.

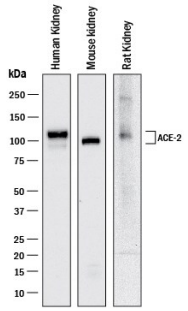
APPLICATIONS

Please Note: *Optimal dilutions should be determined by each laboratory for each application. General Protocols are available in the Technical Information section on our website.*

	Recommended Concentration	Sample
Western Blot	1 µg/mL	See Below
Flow Cytometry	0.25 µg/10^6 cells	See Below
Immunohistochemistry	3-15 µg/mL	See Below
Immunoprecipitation	25 µg/mL	Conditioned cell culture medium spiked with Recombinant Human ACE-2 (Catalog # 933-ZN), see our available Western blot detection antibodies
Simple Western	10 µg/mL	See Below
Blockade of Receptor-ligand Interaction	Hoffman, M. *et al.* (2020) Cell. DOI: 10.1016/j.cell.2020.02.052. This application was not tested by R&D Systems.	

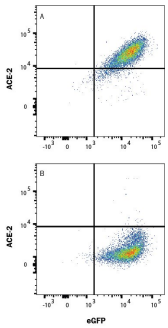
DATA

Western Blot



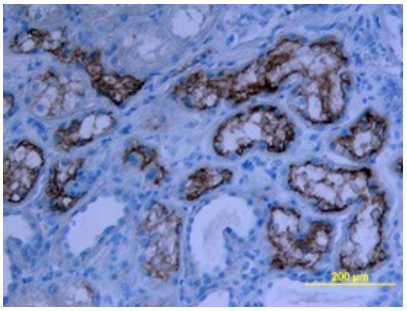
Detection of Human, Mouse, and Rat ACE-2 by Western Blot. Western blot shows lysates of human kidney tissue, mouse kidney tissue, and rat kidney tissue. PVDF membrane was probed with 1 µg/mL of Goat Anti-Human/Mouse/Rat/Hamster ACE-2 Antigen Affinity-purified Polyclonal Antibody (Catalog # AF933) followed by HRP-conjugated Anti-Goat IgG Secondary Antibody (Catalog # Catalog # HAF017). A specific band was detected for ACE-2 at approximately 100 and 110 kDa (as indicated). This experiment was conducted under reducing conditions and using Western Blot Buffer Group 1.

Flow Cytometry



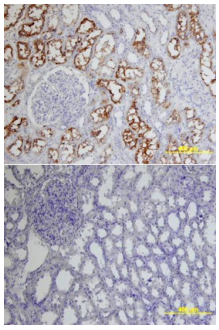
Detection of ACE-2 in HEK293 Human Cell Line Transfected with Human ACE-2 and eGFP by Flow Cytometry. HEK293 human embryonic kidney cell line transfected with (A) human ACE-2 or (B) irrelevant protein, and eGFP was stained with Goat Anti-Human/Mouse/Rat/Hamster ACE-2 Affinity Purified Polyclonal Antibody (Catalog # AF933) followed by Allophycocyanin-conjugated Anti-Goat IgG Secondary Antibody (Catalog # F0108). Quadrant markers were set based on Goat IgG control antibody (Catalog # AB-108-C, data not shown). Staining was performed using our Staining Membrane-associated Proteins protocol.

Immunohistochemistry



ACE-2 in Human Kidney. ACE-2 was detected in immersion fixed paraffin-embedded sections of human kidney using Goat Anti-Human/Mouse/Rat/Hamster ACE-2 Antigen Affinity-purified Polyclonal Antibody (Catalog # AF933) at 15 µg/mL overnight at 4 °C. Tissue was stained using the Anti-Goat HRP-DAB Cell & Tissue Staining Kit (brown; Catalog # Catalog # CTS008) and counterstained with hematoxylin (blue). View our protocol for Chromogenic IHC Staining of Paraffin-embedded Tissue Sections.

Immunohistochemistry



ACE-2 in Human Kidney. ACE-2 was detected in immersion fixed paraffin-embedded sections of human kidney using Goat Anti-Human/Mouse/Rat/Hamster ACE-2 Antigen Affinity-purified Polyclonal Antibody (Catalog # AF933) at 15 µg/mL overnight at 4 °C. Tissue was stained using the Anti-Goat HRP-DAB Cell & Tissue Staining Kit (brown; Catalog # Catalog # CTS008) and counterstained with hematoxylin (blue). Lower panel shows a lack of labeling if primary antibodies are omitted and tissue is stained only with secondary antibody followed by incubation with detection reagents. View our protocol for Chromogenic IHC Staining of Paraffin-embedded Tissue Sections.

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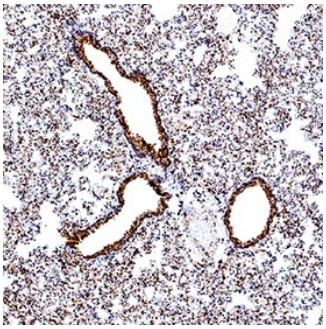
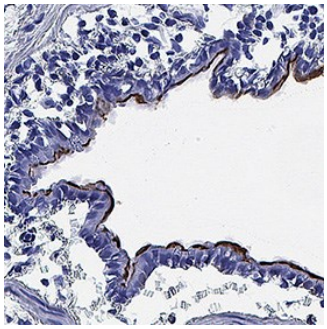
Immunohistochemistry



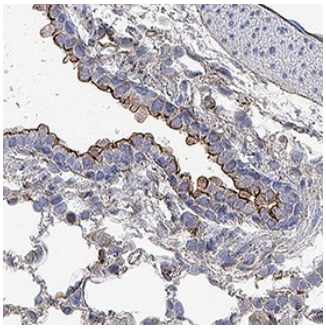
ACE-2 in Hamster Lung. ACE-2 was detected in immersion fixed paraffin-embedded sections of hamster lung using Goat Anti-Human/Mouse/Rat/Hamster ACE-2 Antigen Affinity-purified Polyclonal Antibody (Catalog # AF933) at 3 µg/mL for 1 hour at room temperature followed by incubation with the Anti-Goat IgG VisUCyte™ HRP Polymer Antibody (Catalog # VC004). Before incubation with the primary antibody, tissue was subjected to heat-induced epitope retrieval using Antigen Retrieval Reagent-Basic (Catalog # CTS013). Tissue was stained using DAB (brown) and counterstained with hematoxylin (blue). Specific staining was localized to respiratory bronchioles. Staining was performed using our protocol for IHC Staining with VisUCyte HRP Polymer Detection Reagents.

Immunohistochemistry



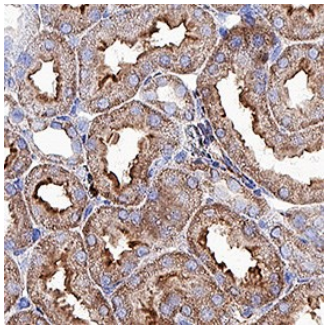
ACE-2 in Rat Lung. ACE-2 was detected in immersion fixed frozen sections of rat lung using Goat Anti-Human/Mouse/Rat/Hamster ACE-2 Antigen Affinity-purified Polyclonal Antibody (Catalog # AF933) at 10 µg/mL for 1 hour at room temperature followed by incubation with the Anti-Goat IgG VisUCyte™ HRP Polymer Antibody (Catalog # VC004). Before incubation with the primary antibody, tissue was subjected to heat-induced epitope retrieval using Antigen Retrieval Reagent-Basic (Catalog # CTS013). Tissue was stained using DAB (brown) and counterstained with hematoxylin (blue). Specific staining was localized to cell surface in epithelial cells in bronchioles. Staining was performed using our protocol for IHC Staining with VisUCyte HRP Polymer Detection Reagents.

Immunohistochemistry



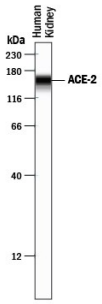
ACE-2 in Rat Lung. ACE-2 was detected in immersion fixed paraffin-embedded sections of rat lung using Goat Anti-Human/Mouse/Rat/Hamster ACE-2 Antigen Affinity-purified Polyclonal Antibody (Catalog # AF933) at 10 µg/mL for 1 hour at room temperature followed by incubation with the Anti-Goat IgG VisUCyte™ HRP Polymer Antibody (Catalog # VC004). Before incubation with the primary antibody, tissue was subjected to heat-induced epitope retrieval using Antigen Retrieval Reagent-Basic (Catalog # CTS013). Tissue was stained using DAB (brown) and counterstained with hematoxylin (blue). Staining was performed using our protocol for IHC Staining with VisUCyte HRP Polymer Detection Reagents.

Immunohistochemistry



ACE-2 in Rat Kidney. ACE-2 was detected in immersion fixed paraffin-embedded sections of rat kidney using Goat Anti-Human/Mouse/Rat/Hamster ACE-2 Antigen Affinity-purified Polyclonal Antibody (Catalog # AF933) at 10 µg/mL for 1 hour at room temperature followed by incubation with the Anti-Goat IgG VisUCyte™ HRP Polymer Antibody (Catalog # VC004). Before incubation with the primary antibody, tissue was subjected to heat-induced epitope retrieval using Antigen Retrieval Reagent-Basic (Catalog # CTS013). Tissue was stained using DAB (brown) and counterstained with hematoxylin (blue). Staining was performed using our protocol for IHC Staining with VisUCyte HRP Polymer Detection Reagents.

Simple Western



Detection of Human ACE-2 by Simple Western™. Simple Western lane view shows lysates of human kidney tissue, loaded at 0.2 mg/mL. A specific band was detected for ACE-2 at approximately 155 kDa (as indicated) using 10 µg/mL of Goat Anti-Human/Mouse/Rat/Hamster ACE-2 Antigen Affinity-purified Polyclonal Antibody (Catalog # AF933) followed by 1:50 dilution of HRP-conjugated Anti-Goat IgG Secondary Antibody (Catalog # Catalog # HAF109). This experiment was conducted under reducing conditions and using the 12-230 kDa separation system.

PREPARATION AND STORAGE

Reconstitution	Reconstitute at 0.2 mg/mL in sterile PBS.
Shipping	The product is shipped at ambient temperature. Upon receipt, store it immediately at the temperature recommended below. *Small pack size (-SP) is shipped with polar packs. Upon receipt, store it immediately at -20 to -70 °C
Stability & Storage	**Use a manual defrost freezer and avoid repeated freeze-thaw cycles.** • 12 months from date of receipt, -20 to -70 °C as supplied. • 1 month, 2 to 8 °C under sterile conditions after reconstitution. • 6 months, -20 to -70 °C under sterile conditions after reconstitution.

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BACKGROUND

ACE-2, also called ACEH (ACE homolog), is an integral membrane protein and a zinc metalloprotease of the ACE family that also includes somatic and germinal ACE (1). Human ACE-2 has about 40% amino acid identity to the N- and C-terminal domains of human somatic ACE. The predicted human ACE-2 protein sequence consists of 805 amino acids, including a N-terminal signal peptide, a single catalytic domain, a C-terminal membrane anchor, and a short cytoplasmic tail. ACE-2 cleaves angiotensins I and II as a carboxypeptidase. ACE-2 mRNA is found at high levels in testis, kidney, and heart and at moderate levels in colon, small intestine, and ovary. Classical ACE inhibitors such as captopril and lisinopril do not inhibit ACE-2 activity. Novel peptide inhibitors of ACE-2 do not inhibit ACE activity (2). Genetic data from Drosophila, mice and rats show that ACE-2 is an essential regulator of heart function in vivo (3).

ACE2 has been shown to be a functional receptor of the human coronaviruses SARS-CoV and SARS-CoV-2 (4, 5). This Human anti-ACE2 antibody (catalog # AF933) was used to block the variant SARS-CoV-2 and ACE2 interaction to elucidate viral transmission and potential therapeutic strategies. (5)

References:

1. Tipnis, S.R. et al. (2000) J. Biol. Chem. **275**:33238.
2. Crackower, M.A. et al. (2002) Nature **417**:822.
3. Huang, L. et al. (2003) J. Biol. Chem. **278**:15532.
4. Li, W. et al. (2003) Nature **426**:450.
5. Hoffmann, M. *et al.* (2020) Cell. DOI: 10.1016/j.cell.2020.02.052.

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